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(54) Title: NOVEL PLANT ACYLTRANSFERASES

(57) Abstract

By this invention, novel nucleic acid sequences encoding for acyltransferase related proteins are provided, wherein said acyltransferase-like protein is active in the transfer of a fatty acyl group from a fatty acyl donor to a fatty acyl acceptor. Also considered are amino acid and nucleic acid sequences obtainable from AT-like nucleic acid sequences and the use of such sequences to provide transgenic host cells capable of producing modified lipid content and composition.

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NOVEL PLANT ACYLTRANSFERASES

5

INTRODUCTION

This application claims the benefit of U.S. Provisional Application Serial No. 60/101,939 filed September 25, 1998.

10

Technical Field

The present invention is directed to nucleic acid and amino acid sequences and constructs, and methods related thereto.

15 Background

Through the development of plant genetic engineering techniques, it is now possible to produce transgenic varieties of plant species to provide plants which have novel and desirable characteristics. For example, it is now possible to genetically engineer plants for tolerance to environmental stresses, such as resistance to pathogens and tolerance to herbicides and to 20 improve the quality characteristics of the plant, for example improved fatty acid compositions. However, the number of useful nucleotide sequences for the engineering of such characteristics is thus far limited and the speed with which new useful nucleotide sequences for engineering new characteristics is slow.

The characterization of various acyltransferase proteins is useful for the further study 25 of plant fatty acid synthesis systems and for the development of novel and/or alternative oils sources. Studies of plant mechanisms may provide means to further enhance, control, modify, or otherwise alter the total fatty acyl composition of triglycerides and oils. Furthermore, the elucidation of the factor(s) critical to the natural production of fatty acids in 30 plants is desired, including the purification of such factors and the characterization of element(s) and/or cofactors which enhance the efficiency of the system. Of particular interest are the nucleic acid sequences of genes encoding proteins which may be useful for applications in genetic engineering.

SUMMARY OF THE INVENTION

5 The present invention provides nucleic acid encoding for amino acid sequences for a class of proteins which are related to acyltransferase proteins. Such proteins are referred to herein as acyltransferase related or acyltransferase like proteins.

By this invention, nucleic acid sequences encoding these acyltransferase related proteins may now be characterized with respect to enzyme activity. In particular,
10 identification and isolation of nucleic acid sequences encoding for acyltransferase related proteins from *Arabidopsis*, yeast, corn, and soybean are provided.

Thus, this invention encompasses acyltransferase related nucleic acid sequences and the corresponding amino acid sequences, and the use of these nucleic acid sequences in the preparation of oligonucleotides containing such acyltransferase related encoding sequences
15 for analysis and recovery of plant acyltransferase related gene sequences. The acyltransferase related encoding sequence may encode a complete or partial sequence depending upon the intended use. All or a portion of the genomic sequence, or cDNA sequence, is intended.

Of special interest are recombinant DNA constructs which provide for transcription or transcription and translation (expression) of the acyltransferase related sequences in host
20 cells. In particular, constructs which are capable of transcription or transcription and translation in plant host cells are preferred. For some applications a reduction in sequences encoding acyltransferase related sequences may be desired. Thus, recombinant constructs may be designed having the acyltransferase related sequences in a reverse orientation for expression of an anti-sense sequence or use of co-suppression, also known as "transwitch",
25 constructs may be useful. Such constructs may contain a variety of regulatory regions including transcriptional initiation regions obtained from genes preferentially expressed in plant seed tissue. For some uses, it may be desired to use the transcriptional and translational initiation regions of the acyltransferase related gene either with the acyltransferase related encoding sequence or to direct the transcription and translation of a heterologous sequence.

30 Also considered in this invention are the plants and seeds containing the constructs and polynucleotides of this invention.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 provides the 204 amino acid conserved sequence profile identified from 5 comparisons of glycerol-3-phosphate acyltransferase and various lysophosphatidic acid acyltransferase using PSI-BLAST.

Figure 2 provides an amino acid sequence alignment for the acyltransferase sequences. The alignment shown is of the regions of the protein extending from about 30 10 amino acids prior to the conserved H in the conserved sequence HXXXXD to 100 amino acids after, or downstream, of the P in the conserved PEG sequence motif of the acyltransferase-like sequences.

Figure 3 provides schematics showing the relationship of the identified acyltransferases. The relationships described are derived from an alignment of the regions of the protein extending from about 30 amino acids prior to the conserved H in the conserved 15 sequence HXXXXD to 100 amino acids after, or downstream, of the P in the conserved PEG sequence motif of the acyltransferase-like sequences. Figure 3A provide a phylogenetic tree showing the relationship of several acyltransferases. Figure 3B provides a table showing the percent similarities and percent divergence of the novel acyltransferases and known acyltransferases using the Clustal method with PAM250 residue weight table.

20

DETAILED DESCRIPTION OF THE INVENTION

In accordance with the subject invention, nucleotide sequences are provided which are 25 capable of coding sequences of amino acids, such as, a protein, polypeptide or peptide, which are related to nucleic acid sequences encoding acyltransferase proteins, referred to herein as acyltransferase-like or acyltransferase related. The novel nucleic acid sequences find use in the preparation of constructs to direct their expression in a host cell. Furthermore, the novel 30 nucleic acid sequences may find use in the preparation of plant expression constructs to modify the fatty acid composition of a plant cell.

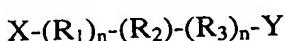
In one embodiment of the present invention, nucleic acid sequences, also referred to herein as polynucleotides, are identified from databases which are related to acyltransferases.

Isolated proteins, Polypeptides and Polynucleotides

A first aspect of the present invention relates to isolated acyltransferase polynucleotides. The polynucleotide sequences of the present invention include isolated 5 polynucleotides that encode the polypeptides of the invention having a deduced amino acid sequence selected from the group of sequences set forth in the Sequence Listing and to other polynucleotide sequences closely related to such sequences and variants thereof.

The invention provides a polynucleotide sequence identical over its entire length to each coding sequence as set forth in the Sequence Listing. The invention also provides the 10 coding sequence for the mature polypeptide or a fragment thereof, as well as the coding sequence for the mature polypeptide or a fragment thereof in a reading frame with other coding sequences, such as those encoding a leader or secretory sequence, a pre-, pro-, or prepro- protein sequence. The polynucleotide can also include non-coding sequences, including for example, but not limited to, non-coding 5' and 3' sequences, such as the 15 transcribed, untranslated sequences, termination signals, ribosome binding sites, sequences that stabilize mRNA, introns, polyadenylation signals, and additional coding sequence that encodes additional amino acids. For example, a marker sequence can be included to facilitate the purification of the fused polypeptide. Polynucleotides of the present invention also include polynucleotides comprising a structural gene and the naturally associated sequences 20 that control gene expression.

The invention also includes polynucleotides of the formula:



wherein, at the 5' end, X is hydrogen, and at the 3' end, Y is hydrogen or a metal, R₁ and R₃ 25 are any nucleic acid residue, n is an integer between 1 and 3000, preferably between 1 and 1000 and R₂ is a nucleic acid sequence of the invention, particularly a nucleic acid sequence selected from the group set forth in the Sequence Listing and preferably SEQ ID NOs: 1, 3, 5, 7, 9, 10, 12, 14, 16, 18, 20, 22, and 226-233. In the formula, R₂ is oriented so that its 5' end residue is at the left, bound to R₁, and its 3' end residue is at the right, bound to R₃. Any 30 stretch of nucleic acid residues denoted by either R group, where R is greater than 1, may be either a heteropolymer or a homopolymer, preferably a heteropolymer.

The invention also relates to variants of the polynucleotides described herein that encode for variants of the polypeptides of the invention. Variants that are fragments of the polynucleotides of the invention can be used to synthesize full-length polynucleotides of the

invention. Preferred embodiments are polynucleotides encoding polypeptide variants wherein 5 to 10, 1 to 5, 1 to 3, 2, 1 or no amino acid residues of a polypeptide sequence of the invention are substituted, added or deleted, in any combination. Particularly preferred are substitutions, additions, and deletions that are silent such that they do not alter the properties 5 or activities of the polynucleotide or polypeptide.

Nucleotide sequences encoding acyltransferases may be obtained from natural sources or be partially or wholly artificially synthesized. They may directly correspond to an acyltransferase endogenous to a natural source or contain modified amino acid sequences, such as sequences which have been mutated, truncated, increased or the like. Acyltransferases 10 may be obtained by a variety of methods, including but not limited to, partial or homogenous purification of protein extracts, protein modeling, nucleic acid probes, antibody preparations and sequence comparisons. Typically an acyltransferase will be derived in whole or in part from a natural source. A natural source includes, but is not limited to, prokaryotic and eukaryotic sources, including, bacteria, yeasts, plants, including algae, and the like.

15 Of special interest are acyltransferases which are obtainable from eukaryotic sources, including those which are obtained, from plants, or from acyltransferases which are obtainable through the use of these sequences. "Obtainable" refers to those acyltransferases which have sufficiently similar sequences to that of the sequences provided herein to provide a biologically active protein of the present invention.

20 Further preferred embodiments of the invention that are at least 50%, 60%, or 70% identical over their entire length to a polynucleotide encoding a polypeptide of the invention, and polynucleotides that are complementary to such polynucleotides. More preferable are polynucleotides that comprise a region that is at least 80% identical over its entire length to a polynucleotide encoding a polypeptide of the invention and polynucleotides that are 25 complementary thereto. In this regard, polynucleotides at least 90% identical over their entire length are particularly preferred, those at least 95% identical are especially preferred. Further, those with at least 97% identity are highly preferred and those with at least 98% and 99% identity are particularly highly preferred, with those at least 99% being the most highly preferred.

30 Preferred embodiments are polynucleotides that encode polypeptides that retain substantially the same biological function or activity as the mature polypeptides encoded by the polynucleotides set forth in the Sequence Listing.

The invention further relates to polynucleotides that hybridize to the above-described sequences. In particular, the invention relates to polynucleotides that hybridize under stringent conditions to the above-described polynucleotides. As used herein, the terms "stringent conditions" and "stringent hybridization conditions" mean that hybridization will generally occur if there is at least 95% and preferably at least 97% identity between the sequences. An example of stringent hybridization conditions is overnight incubation at 42°C, in a solution comprising 50% formamide, 5x SSC (150 mM NaCl, 15 mM trisodium citrate), 50 mM sodium phosphate (pH 7.6), 5x Denhardt's solution, 10% dextran sulfate, and 20 micrograms/milliliter denatured, sheared salmon sperm DNA, followed by washing the hybridization support in 0.1x SSC at approximately 65°C. Other hybridization and wash conditions are well known and are exemplified in Sambrook, *et al.*, Molecular Cloning: A Laboratory Manual, Second Edition, cold Spring Harbor, NY (1989), particularly Chapter 11.

The invention also provides a polynucleotide consisting essentially of a polynucleotide sequence obtainable by screening an appropriate library containing the complete gene for a polynucleotide sequence set forth in the Sequence Listing under stringent hybridization conditions with a probe having the sequence of said polynucleotide sequence or a fragment thereof; and isolating said polynucleotide sequence. Fragments useful for obtaining such a polynucleotide include, for example, probes and primers as described herein.

As discussed herein regarding polynucleotide assays of the invention, for example, polynucleotides of the invention can be used as a hybridization probe for RNA, cDNA, or genomic DNA to isolate full length cDNAs or genomic clones encoding a polypeptide and to isolate cDNA or genomic clones of other genes that have a high sequence similarity to a polynucleotide set forth in the Sequence Listing. Such probes will generally comprise at least 15 bases. Preferably such probes will have at least 30 bases and can have at least 50 bases. Particularly preferred probes will have between 30 bases and 50 bases, inclusive.

The coding region of each gene that comprises or is comprised by a polynucleotide sequence set forth in the Sequence Listing may be isolated by screening using a DNA sequence provided in the Sequence Listing to synthesize an oligonucleotide probe. A labeled oligonucleotide having a sequence complementary to that of a gene of the invention is then used to screen a library of cDNA, genomic DNA or mRNA to identify members of the library which hybridize to the probe. For example, synthetic oligonucleotides are prepared which correspond to the N-terminal sequence of the polypeptide. The partial sequences so prepared can then be used as probes to obtain acyltransferase clones from a gene library prepared from

a cell source of interest. Alternatively, where oligonucleotides of low degeneracy can be prepared from particular peptides, such probes may be used directly to screen gene libraries for gene sequences. In particular, screening of cDNA libraries in phage vectors is useful in such methods due to lower levels of background hybridization.

5 Typically, a sequence obtainable from the use of nucleic acid probes will show 60-70% sequence identity between the target acyltransferase sequence and the encoding sequence used as a probe. However, lengthy sequences with as little as 50-60% sequence identity may also be obtained. The nucleic acid probes may be a lengthy fragment of the nucleic acid sequence, or may also be a shorter, oligonucleotide probe. When longer nucleic acid
10 fragments are employed as probes (greater than about 100 bp), one may screen at lower stringencies in order to obtain sequences from the target sample which have 20-50% deviation (i.e., 50-80% sequence homology) from the sequences used as probe.
Oligonucleotide probes can be considerably shorter than the entire nucleic acid sequence
encoding an acyltransferase enzyme, but should be at least about 10, preferably at least about
15 15, and more preferably at least about 20 nucleotides. A higher degree of sequence identity is
desired when shorter regions are used as opposed to longer regions. It may thus be desirable
to identify regions of highly conserved amino acid sequence to design oligonucleotide probes
for detecting and recovering other related genes. Shorter probes are often particularly useful
20 for polymerase chain reactions (PCR), especially when highly conserved sequences can be
identified. (See, Gould, *et al.*, *PNAS USA* (1989) 86:1934-1938).

25 The skilled artisan will appreciate that, in many cases, an isolated cDNA sequence
will be incomplete, in that the region coding for the polypeptide is truncated with respect to
the 5' terminus of the cDNA. This is a consequence of the reverse transcriptase, an enzyme
with low 'processivity' (a measure of the ability of the enzyme to remain attached to the
template during the polymerization reaction) employed during the first strand cDNA
synthesis.

30 There are several methods available and are well known to the skilled artisan to obtain
full-length cDNAs, or extend short cDNAs, for example those based on the method of Rapid
Amplification of cDNA Ends (RACE) (see, for example, Frohman *et al.* (1988) *Proc. Natl.
Acad. Sci. USA* 85:8998-9002). Recent modifications of the technique, exemplified by the
Marathon™ technology (Clontech Laboratories, Inc.) for example, have significantly
simplified obtaining full-length cDNA sequences.

Another aspect of the present invention relates to isolated acyltransferase polypeptides. Such polypeptides include isolated polypeptides set forth in the Sequence Listing, as well as polypeptides and fragments thereof, particularly those polypeptides which exhibit acyltransferase activity and also those polypeptides which have at least 50%, 60% or 5 70% identity, preferably at least 80% identity, more preferably at least 90% identity, and most preferably at least 95% identity to a polypeptide sequence selected from the group of sequences set forth in the Sequence Listing, and also include portions of such polypeptides, wherein such portion of the polypeptide preferably includes at least 30 amino acids and more preferably includes at least 50 amino acids.

10 "Identity", as is well understood in the art, is a relationship between two or more polypeptide sequences or two or more polynucleotide sequences, as determined by comparing the sequences. In the art, "identity" also means the degree of sequence relatedness between polypeptide or polynucleotide sequences, as determined by the match between strings of such sequences. "Identity" can be readily calculated by known methods including, but not limited 15 to, those described in *Computational Molecular Biology*, Lesk, A.M., ed., Oxford University Press, New York (1988); *Biocomputing: Informatics and Genome Projects*, Smith, D.W., ed., Academic Press, New York, 1993; *Computer Analysis of Sequence Data, Part I*, Griffin, A.M. and Griffin, H.G., eds., Humana Press, New Jersey (1994); *Sequence Analysis in Molecular Biology*, von Heinje, G., Academic Press (1987); *Sequence Analysis Primer*, 20 Gribskov, M. and Devereux, J., eds., Stockton Press, New York (1991); and Carillo, H., and Lipman, D., *SIAM J Applied Math*, 48:1073 (1988). Methods to determine identity are designed to give the largest match between the sequences tested. Moreover, methods to determine identity are codified in publicly available programs. Computer programs which can be used to determine identity between two sequences include, but are not limited to, GCG 25 (Devereux, J., et al., *Nucleic Acids Research* 12(1):387 (1984); suite of five BLAST programs, three designed for nucleotide sequences queries (BLASTN, BLASTX, and TBLASTX) and two designed for protein sequence queries (BLASTP and TBLASTN) (Coulson, *Trends in Biotechnology*, 12: 76-80 (1994); Birren, et al., *Genome Analysis, 1*: 543-559 (1997)). The BLAST X program is publicly available from NCBI and other sources 30 (BLAST Manual, Altschul, S., et al., NCBI NLM NIH, Bethesda, MD 20894; Altschul, S., et al., *J. Mol. Biol.*, 215:403-410 (1990)). The well known Smith Waterman algorithm can also be used to determine identity.

Parameters for polypeptide sequence comparison typically include the following:

Algorithm: Needleman and Wunsch, *J. Mol. Biol.* 48:443-453 (1970)

Comparison matrix: BLOSSUM62 from Hentikoff and Hentikoff, *Proc. Natl. Acad. Sci USA* 89:10915-10919 (1992)

5 Gap Penalty: 12

Gap Length Penalty: 4

A program which can be used with these parameters is publicly available as the "gap" program from Genetics Computer Group, Madison Wisconsin. The above parameters along with no penalty for end gap are the default parameters for peptide comparisons.

10 Parameters for polynucleotide sequence comparison include the following:

Algorithm: Needleman and Wunsch, *J. Mol. Biol.* 48:443-453 (1970)

Comparison matrix: matches = +10; mismatches = 0

Gap Penalty: 50

Gap Length Penalty: 3

15 A program which can be used with these parameters is publicly available as the "gap" program from Genetics Computer Group, Madison Wisconsin. The above parameters are the default parameters for nucleic acid comparisons.

The invention also includes polypeptides of the formula:



20 wherein, at the amino terminus, X is hydrogen, and at the carboxyl terminus, Y is hydrogen or a metal, R₁ and R₃ are any amino acid residue, n is an integer between 1 and 1000, and R₂ is an amino acid sequence of the invention, particularly an amino acid sequence selected from the group set forth in the Sequence Listing and preferably SEQ ID NOs: 2, 4, 6, 8, 11, 13, 15, 17, 19, 21, 23, and 218-225. In the formula, R₂ is oriented so that its amino terminal residue is at the left, bound to R₁, and its carboxy terminal residue is at the right, bound to R₃. Any stretch of amino acid residues denoted by either R group, where R is greater than 1, may be either a heteropolymer or a homopolymer, preferably a heteropolymer.

25 Polypeptides of the present invention include isolated polypeptides encoded by a polynucleotide comprising a sequence selected from the group of a sequence contained in SEQ ID NOs: 1, 3, 5, 7, 9, 10, 12, 14, 16, 18, 20, 22, and 226-233.

30 The polypeptides of the present invention can be mature protein or can be part of a fusion protein.

Fragments and variants of the polypeptides are also considered to be a part of the invention. A fragment is a variant polypeptide which has an amino acid sequence that is entirely the same as part but not all of the amino acid sequence of the previously described polypeptides. The fragments can be "free-standing" or comprised within a larger polypeptide of which the fragment forms a part or a region, most preferably as a single continuous region.

5 Preferred fragments are biologically active fragments which are those fragments that mediate activities of the polypeptides of the invention, including those with similar activity or improved activity or with a decreased activity. Also included are those fragments that are antigenic or immunogenic in an animal, particularly a human.

10 Variants of the polypeptide also include polypeptides that vary from the sequences set forth in the Sequence Listing by conservative amino acid substitutions, substitution of a residue by another with like characteristics. In general, such substitutions are among Ala, Val, Leu and Ile; between Ser and Thr; between Asp and Glu; between Asn and Gln; between Lys and Arg; or between Phe and Tyr. Particularly preferred are variants in which 5 to 10; 1 to 5; 1 to 3 or one amino acid(s) are substituted, deleted, or added, in any combination.

15

Variants that are fragments of the polypeptides of the invention can be used to produce the corresponding full length polypeptide by peptide synthesis. Therefore, these variants can be used as intermediates for producing the full-length polypeptides of the invention.

20 The polynucleotides and polypeptides of the invention can be used, for example, in the transformation of various host cells, as further discussed herein.

The invention also provides polynucleotides that encode a polypeptide that is a mature protein plus additional amino or carboxyl-terminal amino acids, or amino acids within the mature polypeptide (for example, when the mature form of the protein has more than one polypeptide chain). Such sequences can, for example, play a role in the processing of a protein from a precursor to a mature form, allow protein transport, shorten or lengthen protein half-life, or facilitate manipulation of the protein in assays or production. It is contemplated that cellular enzymes can be used to remove any additional amino acids from the mature protein.

25

30 A precursor protein, having the mature form of the polypeptide fused to one or more prosequences may be an inactive form of the polypeptide. The inactive precursors generally are activated when the prosequences are removed. Some or all of the prosequences may be removed prior to activation. Such precursor protein are generally called proproteins.

The polynucleotide and polypeptide sequences can also be used to identify additional sequences which are homologous to the sequences of the present invention. The most preferable and convenient method is to store the sequence in a computer readable medium, for example, floppy disk, CD ROM, hard disk drives, external disk drives and DVD, and then to use the stored sequence to search a sequence database with well known searching tools.

5 Examples of public databases include the DNA Database of Japan
(DDBJ)(<http://www.ddbj.nig.ac.jp/>); Genebank

(<http://www.ncbi.nlm.nih.gov/web/Genbank/Index.html>); and the European Molecular Biology Laboratory Nucleic Acid Sequence Database (EMBL)

10 (http://www.ebi.ac.uk/ebi_docs/embl_db.html). A number of different search algorithms are available to the skilled artisan, one example of which are the suite of programs referred to as BLAST programs. There are five implementations of BLAST, three designed for nucleotide sequences queries (BLASTN, BLASTX, and TBLASTX) and two designed for protein sequence queries (BLASTP and TBLASTN) (Coulson, *Trends in Biotechnology*, 12: 76-80 (1994); Birren, *et al.*, *Genome Analysis*, 1: 543-559 (1997)). Additional programs are available in the art for the analysis of identified sequences, such as sequence alignment programs, programs for the identification of more distantly related sequences, and the like, and are well known to the skilled artisan.

20 **Plant Constructs and Methods of Use**

Of interest in the present invention, is the use of the nucleotide sequences, or polynucleotides, in recombinant DNA constructs to direct the transcription or transcription and translation (expression) of the acyltransferase sequences of the present invention in a host cell.

25 Of particular interest is the use of the nucleotide sequences, or polynucleotides, in recombinant DNA constructs to direct the transcription or transcription and translation (expression) of the acyltransferase sequences of the present invention in a host cell. The expression constructs generally comprise a promoter functional in a host cell operably linked to a nucleic acid sequence encoding an acyltransferase of the present invention and a transcriptional termination region functional in a host cell.

30 By "host cell" is meant a cell which contains a vector and supports the replication, and/or transcription or transcription and translation (expression) of the expression construct.

Host cells for use in the present invention can be prokaryotic cells, such as *E. coli*, or eukaryotic cells such as yeast, plant, insect, amphibian, or mammalian cells. Preferably, host cells are monocotyledenous or dicotyledenous plant cells.

Of particular interest in the present invention is the use of the polynucleotides of the present invention for the preparation of constructs to direct the transcription or transcription and translation of the nucleotide sequences encoding an acyltransferase in a host plant cell. Plant expression constructs generally comprise a promoter functional in a plant host cell operably linked to a nucleic acid sequence of the present and a transcriptional termination region functional in a host plant cell.

Those skilled in the art will recognize that there are a number of promoters which are functional in plant cells, and have been described in the literature. Chloroplast and plastid specific promoters, chloroplast or plastid functional promoters, and chloroplast or plastid operable promoters are also envisioned.

One set of promoters are constitutive promoters such as the CaMV35S or FMV35S promoters that yield high levels of expression in most plant organs. Enhanced or duplicated versions of the CaMV35S and FMV35S promoters are useful in the practice of this invention (Odell, *et al.* (1985) *Nature* 313:810-812; Rogers, U.S. Patent Number 5,378,619). In addition, it may also be preferred to bring about expression of the protein of interest in specific tissues of the plant, such as leaf, stem, root, tuber, seed, fruit, etc., and the promoter chosen should have the desired tissue and developmental specificity.

Of particular interest is the expression of the nucleic acid sequences of the present invention from transcription initiation regions which are preferentially expressed in a plant seed tissue. Examples of such seed preferential transcription initiation sequences include those sequences derived from sequences encoding plant storage protein genes or from genes involved in fatty acid biosynthesis in oilseeds. Examples of such promoters include the 5' regulatory regions from such genes as napin (Kridl *et al.*, *Seed Sci. Res.* 1:209:219 (1991)), phaseolin, zein, soybean trypsin inhibitor, ACP, stearoyl-ACP desaturase, soybean α' subunit of β -conglycinin (soy 7s, (Chen *et al.*, *Proc. Natl. Acad. Sci.*, 83:8560-8564 (1986))) and oleosin.

It may be advantageous to direct the localization of proteins conferring acyltransferase to a particular subcellular compartment, for example, to the mitochondrion, endoplasmic reticulum, vacuoles, chloroplast or other plastidic compartment. For example, where the genes of interest of the present invention will be targeted to plastids, such as chloroplasts, for

expression, the constructs will also employ the use of sequences to direct the gene to the plastid. Such sequences are referred to herein as chloroplast transit peptides (CTP) or plastid transit peptides (PTP). In this manner, where the gene of interest is not directly inserted into the plastid, the expression construct will additionally contain a gene encoding a transit peptide to direct the gene of interest to the plastid. The chloroplast transit peptides may be derived from the gene of interest, or may be derived from a heterologous sequence having a CTP. Such transit peptides are known in the art. See, for example, Von Heijne *et al.* (1991) *Plant Mol. Biol. Rep.* 9:104-126; Clark *et al.* (1989) *J. Biol. Chem.* 264:17544-17550; della-Cioppa *et al.* (1987) *Plant Physiol.* 84:965-968; Romer *et al.* (1993) *Biochem. Biophys. Res Commun.* 196:1414-1421; and, Shah *et al.* (1986) *Science* 233:478-481. Additional transit peptides for the translocation of the protein to the endoplasmic reticulum (ER), or vacuole may also find use in the constructs of the present invention.

Depending upon the intended use, the constructs may contain the nucleic acid sequence which encodes the entire acyltransferase protein, or a portion thereof. For example, where antisense inhibition of a given acyltransferase protein is desired, the entire sequence is not required. Furthermore, where acyltransferase sequences used in constructs are intended for use as probes, it may be advantageous to prepare constructs containing only a particular portion of a acyltransferase encoding sequence, for example a sequence which is discovered to encode a highly conserved acyltransferase region.

The skilled artisan will recognize that there are various methods for the inhibition of expression of endogenous sequences in a host cell. Such methods include, but are not limited to antisense suppression (Smith, *et al.* (1988) *Nature* 334:724-726), co-suppression (Napoli, *et al.* (1989) *Plant Cell* 2:279-289), ribozymes (PCT Publication WO 97/10328), and combinations of sense and antisense, such as those described by Waterhouse, *et al.* (1998) *Proc. Natl. Acad. Sci. USA* 95:13959-13964. Methods for the suppression of endogenous sequences in a host cell typically employ the transcription or transcription and translation of at least a portion of the sequence to be suppressed. Such sequences may be homologous to coding as well as non-coding regions of the endogenous sequence.

Regulatory transcript termination regions may be provided in plant expression constructs of this invention as well. Transcript termination regions may be provided by the DNA sequence encoding the acyltransferase or a convenient transcription termination region derived from a different gene source, for example, the transcript termination region which is naturally associated with the transcript initiation region. The skilled artisan will recognize

that any convenient transcript termination region which is capable of terminating transcription in a plant cell may be employed in the constructs of the present invention.

Alternatively, constructs may be prepared to direct the expression of the acyltransferase sequences directly from the host plant cell plastid. Such constructs and methods are known in the art and are generally described, for example, in Svab, *et al.* (1990) *Proc. Natl. Acad. Sci. USA* 87:8526-8530 and Svab and Maliga (1993) *Proc. Natl. Acad. Sci. USA* 90:913-917 and in U.S. Patent Number 5,693,507.

A plant cell, tissue, organ, or plant into which the recombinant DNA constructs containing the expression constructs have been introduced is considered transformed, transfected, or transgenic. A transgenic or transformed cell or plant also includes progeny of the cell or plant and progeny produced from a breeding program employing such a transgenic plant as a parent in a cross and exhibiting an altered genotype resulting from the presence of an introduced acyltransferase nucleic acid sequence.

The term "introduced" in the context of inserting a nucleic acid sequence into a cell, means "transfection", or "transformation" or "transduction" and includes reference to the incorporation of a nucleic acid sequence into a eukaryotic or prokaryotic cell where the nucleic acid sequence may be incorporated into the genome of the cell (for example, chromosome, plasmid, plastid, or mitochondrial DNA), converted into an autonomous replicon, or transiently expressed (for example, transfected mRNA).

Plant expression or transcription constructs having an acyltransferase as the DNA sequence of interest for increased or decreased expression thereof may be employed with a wide variety of plant life, particularly, plant life involved in the production of vegetable oils for edible and industrial uses. Plants of interest in the present invention include monocotyledenous and dicotyledenous plants. Most especially preferred are temperate oilseed crops. Plants of interest include, but are not limited to, rapeseed (Canola and High Erucic Acid varieties), sunflower, safflower, cotton, soybean, peanut, coconut and oil palms, and corn. Depending on the method for introducing the recombinant constructs into the host cell, other DNA sequences may be required. Importantly, this invention is applicable to dicotyledyons and monocotyledons species alike and will be readily applicable to new and/or improved transformation and regulation techniques.

As used herein, the term "plant" includes reference to whole plants, plant organs (for example, leaves, stems, roots, etc.), seeds, and plant cells and progeny of same. Plant cell, as used herein includes, without limitation, seeds suspension cultures, embryos, meristematic

regions, callus tissue, leaves roots shoots, gametophytes, sporophytes, pollen, and microspores. The class of plants which can be used in the methods of the present invention is generally as broad as the class of higher plants amenable to transformation techniques, including both monocotyledenous and dicotyledenous plants. Particularly preferred plants of 5 interest include, but are not limited to, rapeseed (Canola and High Erucic Acid varieties), sunflower, safflower, cotton, soybean, peanut, coconut and oil palms, and corn. Most especially preferred plants include *Brassica*, soybean, and corn.

As used herein, "transgenic plant" includes reference to a plant which comprises within its genome a heterologous polynucleotide. Generally, the heterologous polynucleotide 10 is stably integrated within the genome such that the polynucleotide is passed on to successive generations. The heterologous polynucleotide may be integrated into the genome alone or as part of a recombinant expression cassette. "Transgenic" is used herein to include any cell, cell line, callus, tissue, plant part or plant, the genotype of which has been altered by the presence of heterologous nucleic acid including those transgenics initially so altered as well 15 as those created by sexual crosses or asexual propagation from the initial transgenic.

Thus a plant having within its cells a heterologous polynucleotide is referred to herein as a transgenic plant. The heterologous polynucleotide can be either stably integrated into the genome, or can be extra-chromosomal. Preferably, the polynucleotide of the present invention is stably integrated into the genome such that the polynucleotide is passed on to 20 successive generations. The polynucleotide is integrated into the genome alone or as part of a recombinant expression cassette. "Transgenic" is used herein to include any cell, cell line, callus, tissue, plant part or plant, the genotype of which has been altered by the presence of heterologous nucleic acids including those transgenics initially so altered as well as those created by sexual crosses or asexual reproduction of the initial transgenics.

As used herein, "heterologous" in reference to a nucleic acid is a nucleic acid that 25 originates from a foreign species, or, if from the same species, is substantially modified from its native form in composition and/or genomic locus by deliberate human intervention. For example, a promoter operably linked to a heterologous structural gene is from a species different from that from which the structural gene was derived, or, if from the same species, 30 one or both are substantially modified from their original form. A heterologous protein may originate from a foreign species, or, if from the same species, is substantially modified from its original form by deliberate human intervention.

As used herein, a "recombinant expression cassette" is a nucleic acid construct, generated recombinantly or synthetically, with a series of specified nucleic acid elements which permit transcription of a particular nucleic acid in a target cell. The recombinant expression cassette can be incorporated into a plasmid, chromosome, mitochondrial DNA, 5 plastid DNA, virus, or nucleic acid fragment. Typically, the recombinant expression cassette portion of an expression vector includes, among other sequences, a nucleic acid sequence to be transcribed and a promoter.

It is contemplated that the gene sequences may be synthesized, either completely or in part, especially where it is desirable to provide plant-preferred sequences. Thus, all or a 10 portion of the desired structural gene (that portion of the gene which encodes the acyltransferase protein) may be synthesized using codons preferred by a selected host. Host-preferred codons may be determined, for example, from the codons used most frequently in the proteins expressed in a desired host species.

One skilled in the art will readily recognize that antibody preparations, nucleic acid 15 probes (DNA and RNA) and the like may be prepared and used to screen and recover "homologous" or "related" acyltransferase from a variety of plant sources. Homologous sequences are found when there is an identity of sequence, which may be determined upon comparison of sequence information, nucleic acid or amino acid, or through hybridization reactions between a known acyltransferase and a candidate source. Conservative changes, 20 such as Glu/Asp, Val/Ile, Ser/Thr, Arg/Lys and Gln/Asn may also be considered in determining sequence homology. Amino acid sequences are considered homologous by as little as 25% sequence identity between the two complete mature proteins. (See generally, Doolittle, R.F., *OF URFS and ORFS* (University Science Books, CA, 1986).)

Thus, other acyltransferase sequences can be obtained from the specific exemplified 25 sequences provided herein. Furthermore, it will be apparent that one can obtain natural and synthetic sequences, including modified amino acid sequences and starting materials for synthetic-protein modeling from the exemplified sequences and from acyltransferases which are obtained through the use of such exemplified sequences. Modified amino acid sequences include sequences which have been mutated, truncated, increased and the like, whether such 30 sequences were partially or wholly synthesized. Sequences which are actually purified from plant preparations or are identical or encode identical proteins thereto, regardless of the method used to obtain the protein or sequence, are equally considered naturally derived.

For immunological screening, antibodies to the acyltransferase protein can be prepared by injecting rabbits or mice with the purified protein or portion thereof, such methods of preparing antibodies being well known to those in the art. Either monoclonal or polyclonal antibodies can be produced, although typically polyclonal antibodies are more useful for gene isolation. Western analysis may be conducted to determine that a related protein is present in a crude extract of the desired plant species, as determined by cross-reaction with the antibodies to the acyltransferase protein. When cross-reactivity is observed, genes encoding the related proteins are isolated by screening expression libraries representing the desired plant species. Expression libraries can be constructed in a variety of commercially available vectors, including lambda gt11, as described in Sambrook, *et al.* (*Molecular Cloning: A Laboratory Manual*, Second Edition (1989) Cold Spring Harbor Laboratory, Cold Spring Harbor, New York).

The nucleic acid sequences associated with acyltransferase proteins will find many uses. For example, recombinant constructs can be prepared which can be used as probes, or which will provide for expression of the acyltransferase protein in host cells to produce a ready source of the enzyme and/or to modify the composition of triglycerides found therein. Other useful applications may be found when the host cell is a plant host cell, either *in vitro* or *in vivo*.

The modification of fatty acid compositions may also affect the fluidity of plant membranes. Different lipid concentrations have been observed in cold-hardened plants, for example. By this invention, one may be capable of introducing traits which will lend to chill tolerance. Constitutive or temperature inducible transcription initiation regulatory control regions may have special applications for such uses.

As discussed above, nucleic acid sequence encoding an acyltransferase of this invention may include genomic, cDNA or mRNA sequence. By "encoding" is meant that the sequence corresponds to a particular amino acid sequence either in a sense or anti-sense orientation. By "extrachromosomal" is meant that the sequence is outside of the plant genome of which it is naturally associated. By "recombinant" is meant that the sequence contains a genetically engineered modification through manipulation via mutagenesis, restriction enzymes, and the like.

Once the desired acyltransferase nucleic acid sequence is obtained, it may be manipulated in a variety of ways. Where the sequence involves non-coding flanking regions, the flanking regions may be subjected to resection, mutagenesis, etc. Thus, transitions,

transversions, deletions, and insertions may be performed on the naturally occurring sequence. In addition, all or part of the sequence may be synthesized. In the structural gene, one or more codons may be modified to provide for a modified amino acid sequence, or one or more codon mutations may be introduced to provide for a convenient restriction site or 5 other purpose involved with construction or expression. The structural gene may be further modified by employing synthetic adapters, linkers to introduce one or more convenient restriction sites, or the like.

The nucleic acid or amino acid sequences encoding an acyltransferase of this invention may be combined with other non-native, or "heterologous", sequences in a variety 10 of ways. By "heterologous" sequences is meant any sequence which is not naturally found joined to the acyltransferase, including, for example, combinations of nucleic acid sequences from the same plant which are not naturally found joined together.

The DNA sequence encoding an acyltransferase of this invention may be employed in conjunction with all or part of the gene sequences normally associated with the 15 acyltransferase. In its component parts, a DNA sequence encoding acyltransferase is combined in a DNA construct having, in the 5' to 3' direction of transcription, a transcription initiation control region capable of promoting transcription and translation in a host cell, the DNA sequence encoding plant acyltransferase and a transcription and translation termination region.

Potential host cells include both prokaryotic cells, such as *E.coli* and eukaryotic cells 20 such as yeast, insect, amphibian, or mammalian cells. A host cell may be unicellular or found in a multicellular differentiated or undifferentiated organism depending upon the intended use. Preferably, host cells of the present invention include plant cells, both 25 monocotyledenous and dicotyledenous. Cells of this invention may be distinguished by having a sequence foreign to the wild-type cell present therein, for example, by having a recombinant nucleic acid construct encoding an acyltransferase therein.

The methods used for the transformation of the host plant cell are not critical to the present invention. The transformation of the plant is preferably permanent, i.e. by integration 30 of the introduced expression constructs into the host plant genome, so that the introduced constructs are passed onto successive plant generations. The skilled artisan will recognize that a wide variety of transformation techniques exist in the art, and new techniques are continually becoming available. Any technique that is suitable for the target host plant can be employed within the scope of the present invention. For example, the constructs can be

introduced in a variety of forms including, but not limited to as a strand of DNA, in a plasmid, or in an artificial chromosome. The introduction of the constructs into the target plant cells can be accomplished by a variety of techniques, including, but not limited to calcium-phosphate-DNA co-precipitation, electroporation, microinjection, *Agrobacterium* infection, liposomes or microprojectile transformation. The skilled artisan can refer to the literature for details and select suitable techniques for use in the methods of the present invention.

Normally, included with the DNA construct will be a structural gene having the necessary regulatory regions for expression in a host and providing for selection of transformant cells. The gene may provide for resistance to a cytotoxic agent, e.g. antibiotic, heavy metal, toxin, etc., complementation providing prototrophy to an auxotrophic host, viral immunity or the like. Depending upon the number of different host species the expression construct or components thereof are introduced, one or more markers may be employed, where different conditions for selection are used for the different hosts.

Where *Agrobacterium* is used for plant cell transformation, a vector may be used which may be introduced into the *Agrobacterium* host for homologous recombination with T-DNA or the Ti- or Ri-plasmid present in the *Agrobacterium* host. The Ti- or Ri-plasmid containing the T-DNA for recombination may be armed (capable of causing gall formation) or disarmed (incapable of causing gall formation), the latter being permissible, so long as the *vir* genes are present in the transformed *Agrobacterium* host. The armed plasmid can give a mixture of normal plant cells and gall.

In some instances where *Agrobacterium* is used as the vehicle for transforming host plant cells, the expression or transcription construct bordered by the T-DNA border region(s) will be inserted into a broad host range vector capable of replication in *E. coli* and *Agrobacterium*, there being broad host range vectors described in the literature. Commonly used is pRK2 or derivatives thereof. See, for example, Ditta, *et al.*, (*Proc. Nat. Acad. Sci., U.S.A.* (1980) 77:7347-7351) and EPA 0 120 515, which are incorporated herein by reference. Alternatively, one may insert the sequences to be expressed in plant cells into a vector containing separate replication sequences, one of which stabilizes the vector in *E. coli*, and the other in *Agrobacterium*. See, for example, McBride and Summerfelt (*Plant Mol. Biol.* (1990) 14:269-276), wherein the pRiHRI (Jouanin, *et al.*, *Mol. Gen. Genet.* (1985) 201:370-374) origin of replication is utilized and provides for added stability of the plant expression vectors in host *Agrobacterium* cells.

Included with the expression construct and the T-DNA will be one or more markers, which allow for selection of transformed Agrobacterium and transformed plant cells. A number of markers have been developed for use with plant cells, such as resistance to chloramphenicol, kanamycin, the aminoglycoside G418, hygromycin, or the like. The 5 particular marker employed is not essential to this invention, one or another marker being preferred depending on the particular host and the manner of construction.

For transformation of plant cells using *Agrobacterium*, explants may be combined and incubated with the transformed *Agrobacterium* for sufficient time for transformation, the bacteria killed, and the plant cells cultured in an appropriate selective medium. Once callus 10 forms, shoot formation can be encouraged by employing the appropriate plant hormones in accordance with known methods and the shoots transferred to rooting medium for regeneration of plants. The plants may then be grown to seed and the seed used to establish repetitive generations and for isolation of vegetable oils.

There are several possible ways to obtain the plant cells of this invention which 15 contain multiple expression constructs. Any means for producing a plant comprising a construct having a nucleic acid sequence of the present invention, and at least one other construct having another DNA sequence encoding an enzyme are encompassed by the present invention. For example, the expression construct can be used to transform a plant at the same time as the second construct either by inclusion of both expression constructs in a single 20 transformation vector or by using separate vectors, each of which express desired genes. The second construct can be introduced into a plant which has already been transformed with the first expression construct, or alternatively, transformed plants, one having the first construct and one having the second construct, can be crossed to bring the constructs together in the same plant.

25 In general, acyltransferase proteins are active in the transfer of acyl groups from a donor to a variety of different substrates. For example, diacylglycerol acyltransferases add acyl groups to diacylglycerol to form triacylglycerol (TAG), or acyl:CoA:cholesterol acyltransferase uses an acyl-CoA as a donor to transfer an acyl group to a sterol to form a sterol ester. Typically, the substrates include, but are not limited to glycerides, including 30 mono and diglycerides, sterols, stanols, phosphatides, and the like. Donors include, but are not limited to acyl-CoA and acyl-ACP molecules.

The invention now being generally described, it will be more readily understood by reference to the following examples which are included for purposes of illustration only and are not intended to limit the present invention.

5

EXAMPLES

Example 1: RNA Isolations

10 Total RNA from the inflorescence and developing seeds of *Arabidopsis thaliana* is isolated for use in construction of complementary (cDNA) libraries. The procedure is an adaptation of the DNA isolation protocol of Webb and Knapp (D.M. Webb and S.J. Knapp, 1990) Plant Molec. Reporter, 8, 180-185). The following description assumes the use of 1g fresh weight of tissue. Frozen seed tissue is powdered by grinding under liquid nitrogen. The
15 powder is added to 10ml REC buffer (50mM Tris-HCl, pH 9, 0.8M NaCl, 10mM EDTA, 0.5% w/v CTAB (cetyltrimethyl-ammonium bromide)) along with 0.2g insoluble polyvinylpolypyrrolidone, and ground at room temperature. The homogenate is centrifuged for 5 minutes at 12,000 xg to pellet insoluble material. The resulting supernatant fraction is extracted with chloroform, and the top phase is recovered.

20 The RNA is then precipitated by addition of 1 volume RecP (50mM Tris-HCL pH9, 10mM EDTA and 0.5% (w/v) CTAB) and collected by brief centrifugation as before. The RNA pellet is redissolved in 0.4 ml of 1M NaCl. The RNA pellet is redissolved in water and extracted with phenol/chloroform. Sufficient 3M potassium acetate (pH 5) is added to make the mixture 0.3M in acetate, followed by addition of two volumes of ethanol to precipitate the
25 RNA. After washing with ethanol, this final RNA precipitate is dissolved in water and stored frozen.

Alternatively, total RNA may be obtained using TRIzol reagent (BRL-Lifetechnologies, Gaithersburg, MD) following the manufacturers protocol. The RNA precipitate is dissolved in water and stored frozen.

30

Example 2: Identification of Acyltransferase Homology Sequences

Searches are performed on a Silicon Graphics Unix computer using additional Bioaccelerator hardware and GenWeb software supplied by Compugen Ltd. This software and hardware enables the use of the Smith-Waterman algorithm in searching DNA and protein databases using profiles as queries. The program used to query protein databases is profilesearch. This is a search where the query is not a single sequence but a profile based on a multiple alignment of amino acid or nucleic acid sequences. The profile is used to query a sequence data set, i.e., a sequence database. The profile contains all the pertinent information for scoring each position in a sequence, in effect replacing the "scoring matrix" used for the standard query searches. The program used to query nucleotide databases with a protein profile is tprofilesearch. Tprofilesearch searches nucleic acid databases using an amino acid profile query. As the search is running, sequences in the database are translated to amino acid sequences in six reading frames. The output file for tprofilesearch is identical to the output file for profilesearch except for an additional column that indicates the frame in which the best alignment occurred.

The Smith-Waterman algorithm, (Smith and Waterman (1981) *supra*), is used to search for similarities between one sequence from the query and a group of sequences contained in the database. E score values as well as other sequence information, such as conserved peptide sequences of HXXXXD and PEG are used to identify related sequences. By using the conserved peptide sequence information, E score values of greater than E-12 and E-8 are considered. For example, the EST sequence originally used to identify ATAT2 had an E score of 0.0094, while the EST sequence originally used to identify ATLPAAT1 had an E score of 0.0868.

A protein sequence of glycerol-3-phosphate from *E. coli* (Swiss Prot Accession P00482) is used to search the NCBI non-redundant protein database using BLAST. In the first round of searches, other membrane forms of G3PAAT are identified. In subsequent PSI-BLAST searches (Altschul, *et al.* (1997) *Nucleic Acids Res* 25:3389-3402), LPAATs and other acyltransferases are identified. Using sequence alignment software programs, G3PAAT and different LPAAT amino acid sequences are aligned, and a profile is generated using a homologous sequence region, between amino acids 256 and 459 of the *E. coli* sequence.

The identified 204 amino acid is used to query the protein database using PSI-BLAST. After 5 iterations of PSI-BLAST, the profile generated from this new query (Figure 1)

identified soluble forms of G3PAAT. Prior to this identification, no sequence homology had been identified between the membrane and soluble forms of G3PAAT.

5 **Example 3: Excision of PSI-BLAST Profile**

The profile generated from the queries using PSI-BLAST is excised from the hyper text markup language (html) file. The worldwide web (www)/html interface to psiblast at ncbi stores the current generated profile matrix in a hidden field in the html file that is 10 returned after each iteration of psiblast. However, this matrix has been encoded into string62 (s62) format for ease of transport through html. String62 format is a simple conversion of the values of the matrix into html legal ascii characters.

The encoded matrix width (x axis) is 26 characters, and comprise the consensus characters, the probabilities of each amino acid in the order A,B,C,D,E,F,G,H,I,K,L,M,N, 15 P,Q,R,S,T,V,W,X,Y,Z (where B represents D and N, and Z represents Q and E, and X represents any amino acid), gap creation value, and gap extension value.

The length (y axis) of the matrix corresponds to the length of the sequences identified by PSI-BLAST. The order of the amino acids corresponds to the conserved amino acid sequence of the sequences identified using PSI-BLAST, with the N-terminal end at the top of 20 the matrix. The probabilities of other amino acids at that position are represented for each amino acid along the x axis, below the respective single letter amino acid abbreviation.

Thus, each row of the profile consists of the highest scoring (consensus) amino acid, followed by the scores for each possible amino acid at that position in sequence matrix, the score for opening a gap at that position, and the score for continuing a gap at that position.

25 The string62 file is converted back into a profile for use in subsequent searches. The gap open field is set to 11 and the gap extension field is set to 1 along the x axis. The gap creation and gap extension values are known, based on the settings given to the PSI-BLAST algorithm. The matrix is exported to the standard GCG profile form. This format can be read by GenWeb.

30 The algorithm used to convert the string62 formatted file to the matrix is outlined in Table 1.

Table 1

1. if encoded character z then the value is blast score min
 2. if encoded character Z then the value is blast score max
 5 3. else if the encoded character is uppercase then its value is (64-(ascii # of char))
 4. else if the encoded character is a digit the value is ((ascii # of char)-48)
 5. else if the encoded character is not uppercase then the value is ((ascii # of char) - 87)
 6. ALL B positions are set to min of D and N amino acids at that row in sequence matrix
 7. ALL Z positions are set to min of Q and E amino acids at that row in sequence matrix
 10 8. ALL X positions are set to min of all amino acids at that row in sequence matrix
 9. kBLAST_SCORE_MAX=999;
 10. kBLAST_SCORE_MIN=-999;
 11. all gap opens are set to 11
 12. all gap lens are set to 1

15

Example 4: Identification of Novel Acyltransferase Related Amino Acid Sequences

20 The profile (Figure 1) is used in further queries to identify a number of previously unidentified proteins from yeast as novel acyltransferases. A protein is identified from an *Arabidopsis* protein sequence database (ATAT1) (SEQ ID NO:2). Sequences are also identified from nucleic acid databases (Table 2)

25

Table 2

Database ID Number	BLAST Search Hits	Log probability
<u><i>Saccharomyces cerevisiae</i></u>		
gi 1078509 NO:217)	Limnanthes putative LPAAT	e-10 (SEQ ID
30 gi 586485 NO:218)	Limnanthes putative LPAAT	e-13 (SEQ ID

gi 320748 NO:219)	Limnanthes putative LPAAT	e-19 (SEQ ID
gi 2506920	SUPPRESSES CTR1 (choline transport mutant) (SEQ ID NO:220)	
gi 549627 5 NO:221)	similar to CTR1	e-118 (SEQ ID
gi 2133031 NO:222)	unidentified	(SEQ ID
gi 2132939 NO:223)	unidentified	(SEQ ID
10 gi 2132299 NO:224)	TAFAZZIN	e-14 (SEQ ID

In Table 2, the gi number is the database identifier, the middle column shows the results of BLAST searches against the NCBI NR protein database, and the log probability number shows represents the log of the probability of such a match occurring by random chance. These proteins, including the ATAT1 protein sequence, are identified using the original PSI-BLAST search of the NCBI NR protein database. Thus, these proteins are novel acyltransferase related proteins with unidentified activities.

The *Arabidopsis* acyltransferase sequence, herein referred to as ATAT1, is also identified using the original PSI-BLAST search of the NCBI NR protein database, and did not have an annotated function.

Additional *Arabidopsis* amino acid sequences related to acyltransferases are identified from the databases, referred to as ATAT2est, ATAT3est, ATAT4est, ATAT5est, ATAT6est, ATAT7est, ATAT8est, ATAT9, ATAT10, and ATAT11est. Furthermore, *Arabidopsis* 25 amino acid sequences are identified which demonstrate sequence similarity to known lysophosphatidic acid, referred to as ATLPAAT1. The sequences of ATAT9 and ATAT10 are identified from the database as genomic sequences, all other *Arabidopsis* sequences are identified as ESTs.

Example 5: Sequence Analysis of the Novel Acyltransferases

To obtain the entire coding region corresponding to the *Arabidopsis* acyltransferase sequences, synthetic oligo-nucleotide primers are designed to amplify the 5' and 3' ends of partial cDNA clones containing acyltransferase related sequences. Primers are designed according to the respective *Arabidopsis* acyltransferase related sequences (Table 3) and used 5 in Rapid Amplification of cDNA Ends (RACE) reactions (Frohman *et al.* (1988) *Proc. Natl. Acad. Sci. USA* 85:8998-9002) using the Marathon cDNA amplification kit (Clontech Laboratories Inc, Palo Alto, CA). Primers with an R designation are used for 5' RACE reactions, and primers with an F designation are used for 3' RACE reactions.

Table 3

	<u>ATAT2</u>	
	ATAT2R1	CCATCCGCTTCAAGGGAACGACACCCATCA (SEQ ID NO:135)
5	ATAT2R2	TCCCTGTCTGCTTGATGAACTTAAAGCTTG (SEQ ID NO:136)
	ATAT2R3	ACAGCAGGAGTGTCTGATGATGGCAGATTG (SEQ ID NO:137)
	<u>ATAT3</u>	
	ATAT3R1	ACTGGAGTTCCAGCCAAAAATGCACCTGTC (SEQ ID NO:138)
10	ATAT3R2	GATACACCCTTGAAATCAGGCGATTTGCT (SEQ ID NO:139)
	<u>ATAT4</u>	
	ATAT4R1	TTGCAAATTCAATT CCTGTTCACCGGGCC (SEQ ID NO:140)
	ATAT4R2	GT T T C T G C T A T T C C A G A A G G C G T C A A C A A (SEQ ID NO:141)
15	<u>ATAT5</u>	
	ATAT5R1	CATTGAAGATCCGTCCGTGAAGTTNCCTTACC (SEQ ID NO:142)
	ATAT5R2	TCGAGCTGTGATCGATGATTGGCTGTGAAG (SEQ ID NO:143)
	ATAT5F1	GTCTCTTCAAAAACACACACACACACGTCTCT (SEQ ID NO:144)
	ATAT5F2	GTCTCTTCAAAAACACACACACACACACGTCTCT (SEQ ID NO:145)
20	<u>ATAT6</u>	
	H76348-F1	GTAGAGAGCCTTACTTGCTTCGGTTAGTC (SEQ ID NO:146)
	H76348-F2	ACGTCATCGTACCTGTTGCTATTGACTCAC (SEQ ID NO:147)
	H76348-R1	ACTTTCCATTGTCAGGGACTCCTCGACAC (SEQ ID NO:148)
25	H76348-R2	ACGGTGTAGGAAGGGAAAGGATTCAAAAGG (SEQ ID NO:149)
	<u>ATAT7</u>	
	ATTS0193-F1	GCGATGA ACTACAGAGTCGGATTCTTCCTC (SEQ ID NO:150)
	ATTS0193-F2	CCGGTTACGAGATTACGTTCTGAACCAAG (SEQ ID NO:151)
30	ATTS0193-R1	CAATGGAGACAAGGCTCGAAAGTGCTAACCC (SEQ ID NO:152)
	ATTS0193-R2	ATTCTCTGAACATAGTTGCCACGGTCATG (SEQ ID NO:153)

ATAT8

AA042618-F1 GAAATCCAACGCCCTCCCAATATCACTCTG (SEQ ID NO:154)

AA042618-F2 CTTCAACTTCCATCAGGATCTTGGCACGT (SEQ ID NO:155)

AA042618-R1 ACCACTTGTAGAGACCTTACCTGCTTAGG (SEQ ID NO:156)

5 AA042618-R2 TCCTACCTACACCATCCAATTCTCGACCC (SEQ ID NO:157)

ATAT11

ATAT11R1 CTGCGTCAAGTGAGCAACTCAGTTCTTGCA (SEQ ID NO:158)

ATAT11R2 TGGGAAGCAGCACGTTGTCAGTATCGGAA (SEQ ID NO:159)

10 ATAT11R3 TAGCCTCTGTGTAATCTGTGCCCTGGGG (SEQ ID NO:160)

From the nucleic acid sequences obtained from the RACE reactions, protein sequence is predicted for each nucleic acid sequence using Macvector software. Nucleic acid sequences 15 are provided for ATAT1 (SEQ ID NO:1), ATAT2 (SEQ ID NO:3), ATAT3 (SEQ ID NO:5), ATAT4 (SEQ ID NO:7), ATAT5 (SEQ ID NO:9), ATAT6 (SEQ ID NO:10), ATAT7 (SEQ ID NO:12), ATAT8 (SEQ ID NO:14), ATAT9 (SEQ ID NO:16), ATAT10 (SEQ ID NO:18), ATAT11 (SEQ ID NO:20) and ATLPAAT1 (SEQ ID NO:22), respectively.

The protein sequence derived from the ATAT1 (SEQ ID NO:2) nucleic acid sequence 20 from Arabidopsis has a predicted molecular mass of 32.5 kDa, and a PI of 9.74. Alignment of the Arabidopsis acyltransferase with several LPAAT and G3PAAT shows that some of the domains that are conserved between LPAAT and G3PAAT are conserved in the new acyltransferase protein.

The ATAT2 nucleic acid sequence is predicted to encode a 312 amino acid protein 25 (SEQ ID NO:4), with a molecular weight of 34.6 kD, and a pI of 9.99. The ATAT2 protein may also contain 2 to 3 transmembrane domains. However, the protein encoded by the ATAT2 nucleic acid sequence may be longer than predicted because of the absence of an inframe stop codon upstream of the ATG start codon used.

The ATAT3 nucleic acid sequence is predicted to encode a 398 amino acid protein 30 (SEQ ID NO:6), with a molecular weight of 44.7 kD, and a pI of 5.62. The ATAT3 protein may contain 1 to 4 transmembrane domains. The ATAT4 nucleic acid sequence is predicted to encode a 317 amino acid protein (SEQ ID NO:8), with a molecular weight of 36.5 kD, and a pI of 9.67. The ATAT4 protein is predicted to have 2 to 5 transmembrane domains.

The ATLPAAT1 nucleic acid sequence is predicted to encode a 389 amino acid protein (SEQ ID NO:23), with a molecular weight of 43.7 kD, and a pI of 9.52. The ATLPAAT1 protein is predicted to have up to 3 transmembrane domains. The protein predicted from the ATLPAAT1 nucleic acid sequence is similar to LPAATs reported for 5 *Brassica*, maize, and meadowfoam (described in PCT Publication WO 94/13814). The ATAT11 nucleic acid sequence is predicted to encode a 375 amino acid protein (SEQ ID NO:21), with a molecular weight of 43.5 kD, and a pI of 9.45. The deduced amino acid sequences of ATAT6 (SEQ ID NO:11), ATAT7 (SEQ ID NO:13), ATAT8 (SEQ ID NO:15), ATAT9 (SEQ ID NO:17), and ATAT10 (SEQ ID NO:19) are also provided

10 A sequence region approximately 30 amino acids upstream through approximately 100 amino acids downstream of the conserved amino acid sequences HXXXXD (Heath and Rock, (1998) *J. Bacteriol.* 180(6):1425-1430) and PEG (Neuwald (1997) *Curr Biol* 7:R465-R466) of the predicted amino acid sequences derived from the nucleic acid sequences of ATAT1, ATAT2, ATAT3, ATAT4, ATAT6, ATAT7, ATAT8, ATAT9, ATAT10, 15 ATLPAAT1, and ATAT11 are compared to the amino acid sequences of lysophosphatidic acid acyltransferase (Jojoba AT (SEQ ID NO:162, the nucleic acid sequence is provided in SEQ ID NO:161), maize AT (PCT Publication WO 94/13814), PLSC coco(GenBank accession 1098605), PLSC Lim(GenBank accession 1209507), PLSC, Ecoli (GenBank accession 1209507), and PLSC Yeast(GenBank accession 464422)) and glycerol-3-phosphate 20 acyltransferase (PLSB Ecoli(GenBank accession 130326) and PLSB Mouse(GenBank accession 2498786)) (Figure 2), and similarities are identified (Figure 2 and Figure 3).

Sequence comparisons reveal several classes of acyltransferases exist based on 25 conserved amino acid sequences identified in the comparisons in Figure 2. For example, ATAT1, ATAT6, ATAT7, ATAT8, and ATAT9, contain the conserved amino acid sequences of VTYXSX(SEQ ID NO: 128), VXLTRXR(SEQ ID NO: 129), LXXGDLV(SEQ ID NO: 132) between the HXXXXD and PEG sequences. In addition, ATAT1, ATAT6, ATAT7, ATAT8, and ATAT9 also contain the conserved sequences CPEGT(SEQ ID NO: 130) which comprises the PEG sequence, as well as IVPVA(SEQ ID NO: 131) and VANXXQ (SEQ ID NO: 134)(Figure 2) downstream of the PEG sequence. The sequences corresponding to ATAT1, ATAT7, and ATAT9 are the most closely related in this class, with 30 similarities between ATAT1 and ATAT9 of 67.0%, between ATAT1 and ATAT7 of 58.2% and between ATAT9 and ATAT7 of 63.9% (Figure 3B).

Sequence comparisons also demonstrate that the sequence of ATLPAAT1 is most closely related to the jojoba LPAAT (82.3% similar), and maize (78.0% similar).

Furthermore, sequence analysis demonstrates that ATAT4 is the most divergent sequence with the highest similarity to ATAT10 (18.5%). The highest similarity (15.3%) to a 5 known sequence is with a meadowfoam (*Limnanthes douglassi*) LPAAT. However, the sequences of ATAT4 and ATAT10 share several conserved peptide sequences with the amino acid sequences of ATAT2 and ATAT3 (Figure 2), VXNHXS (SEQ ID NO: 127) where the H comprises the conserved H of the HXXXXD sequence and FXXGAF (SEQ ID NO: 133) downstream of the PEG sequence.

10

Example 6: Identification of Additional Acyltransferase Sequences

The novel *Arabidopsis* sequences identified above are used to search proprietary 15 databases containing soybean and corn EST sequences. The results of this search identifies EST sequences from soybean (SEQ ID NO:24 through SEQ ID NO: 85) as well as from corn (SEQ ID NO: 86 through SEQ ID NO:126) as encoding acyltransferase related proteins.

Sequence comparisons between the various EST sequences and the complete 20 *Arabidopsis* sequences reveals that the identified EST sequences demonstrate higher similarity to the various *Arabidopsis* sequences as determined by BLAST scores.

Expressed Sequence Tag (EST) sequences from soybean and corn databases are identified which are most closely related by BLAST score to ATAT1 (SEQ ID NOS:24-29 and SEQ ID NOS:86-88, respectively), ATAT2 (SEQ ID NO: 30 and SEQ ID NO:89, respectively), ATAT3 (SEQ ID NOS:31-35 and SEQ ID NOS:90-94, respectively), ATAT4 25 (SEQ ID NOS:36-44 and SEQ ID NOS:95-100, respectively), ATAT6 (SEQ ID NOS:45-49 and SEQ ID NO:101, respectively), ATAT7 (SEQ ID NOS:50-54 and SEQ ID NOS:102-103, respectively), ATAT8 (SEQ ID NOS:55-56 and SEQ ID NO:104, respectively), ATAT9 (SEQ ID NOS:57-79 and SEQ ID NOS:105-111, respectively), ATAT10 (SEQ ID NOS:80-81 and SEQ ID NO:112, respectively), ATAT11, (SEQ ID NOS:82-85 and SEQ ID 30 NOS:123-126, respectively), and ATLPAAT1 (SEQ ID NOS: 113-122 respectively).

Example 7: Expression Construct Preparation

A series of synthetic oligo nucleotide primers were prepared for use in Polymerase Chain Reactions (PCR) to amplify the entire DNA sequences encoding the various acyltransferase sequences identified above. The sequences are listed in Table 3.

Table 3

Primer	Sequence (listed 5'-3')	SEQ ID NO:
ATAT1F	AAGCTTGCATGCGTCGACACAATGGTCATGCGACCAAGT CAG	163
ATAT1R	GGTACCGTCGACTCACTTCTTGGTGTGTTGATAG	164
ATAT2F	GGATCCGGGCCGACAATGACGAGCTTACTACTTCCCT TCAT	165
ATAT2R	GGATCCCCTGCAGGTTAGAGATCCATTGATTCTGCAAT	166
ATAT3F	GGATCCGGGCCGCGATAATGGAATCAGAGCTAAAGAT	167
ATAT3R	GGATCCCCTGCAGGTCAATTCTCTTGATGGAAATC	168
ATAT4F	GGATCCGGGCCGACAATGACTCGTTACAAGATGTTTC A	169
ATAT4R	GGATCCCCTGCAGGTCACTTCTCTTCCAATCTAGCCAG	170
ATAT6F	GGATCCGGGCCGCGACAATGTCCGGTAATAAGATCTGAC TCTTCA	171
ATAT6R	GGATCCCCTGCAGGTTATTTTCTTGACAACCTCCGTTAT TACCGG	172
ATAT7F	ATATCCGGGCCGACAATGGTTATGGAGCAAGCTGGAA	173
ATAT7R	GGATCCCCTGCAGGTCAATGGAGACAAGGCTCGAAAGT	174
ATAT8F	GGATCCGGGCCGACAATGTCCGCCAAGATTCATATT CC	175
ATAT8R	GGATCCCCTGCAGGTTAATTTCTTAACACTCCATT	176
ATAT9F	GGATCCGGGCCGACAATGGGAGCTCAGGAGAACGGCG CC	177
ATAT9R	GGATCCCCTGCAGGTACAGTCTTCTCCTTCTTACCGG	178
ATAT10F	GGATCCGGGCCGACAATGGCGGATCCTGATCTGTCTTC TCCT	179
ATAT10R	GGATCCCCTGCAGGTTATGTTGGGCCAAGTCAGGTGCAA AGAT	180
ATAT11F	GGATCCGGGCCGAAAATGGAAAAAAAGAGTGTACCAAA	181

	TTCT	
ATAT11R	GGATCCCCTGCAGGTTATTGTTACTAATTGAGGAAAT	182
	TTTTG	
ATLPAAT	TCGACCTGCAGGAAGCTTAAGGATGGTGATTGCTGC	183
1F		
ATLPAAT	GGATCCCGGCCGCTTACTTCCTCCTCTCCG	184
1R		
YSCAT1F	GGATCCCGGCCGACAATGTCTTTAGGGATGTCCTAG	185
YSCAT1R	GGATCCCCTGCAGGTCAATCATCCTTACCCCTTGGTTAC	186
C		
YSCAT 1	ATGTCTTTAGGGATGTCCTAGAAAGAGGAGATGAATT	187
KO F	CTGTGCGGTATTCACACCG	
YSCAT 1	TCAATCATCCTTACCCCTTGGTTACCCTCTGGAGGCAGA	188
KO R	AGATTGTACTGAGAGTGCAC	
YSCAT2F	GGATCCCGGCCGACAATGAAGCATTCCAAAAATACCG	189
	TAGG	
YSCAT2R	GGATCCCCTGCAGGTCAATGATTTCATCACAAATA	190
C		
YSCAT 2	ATGAAGCATTCCAAAAATACCGTAGGTATGGAATTATG	191
KO F	CTGTGCGGTATTCACACCG	
YSCAT 2	TCAATGATTTTCATCACAAATAAGAATAAGAAAA	192
KO R	AGATTGTACTGAGAGTGCAC	
YSCAT	GGATCCCGGCCGACAATGGGTTTGATTCTTCGA	193
3F	AAC	
YSCAT	GGATCCCCTGCAGGTTATTGGTCTCAATTAAATATT	194
3R	TTTGC	
YSCAT 3	ATGGGTTTGTTGATTCTCGAACATATATGGTCGGTT	195
KO F	CTGTGCGGTATTCACACCG	
YSCAT 3	TTATTTGGTCTCAATTAAATATTTTGCAAGGACTCG	196
KO R	AGATTGTACTGAGAGTGCAC	
YSCAT	GGATCCCGGCCGACAATGGAAAAGTACACCAATTGGAG	197
4F	AGAC	
YSCAT	GGATCCCCTGCAGGCTACTTCCTTTACGTTGATCGC	198
4R	TG	
YSCAT 4	ATGGAAAAGTACACCAATTGGAGAGACAATGGTACGGAA	199
KO F	CTGTGCGGTATTCACACCG	
YSCAT 4	CTACTTCCTTTACGTTGATCGCTGATATATTCTTC	200
KO R	AGATTGTACTGAGAGTGCAC	

YSCAT	GGATCCGGCGGCCGCACAATGCCTGCACCAAAACTCACGGA	201
5F	G	
YSCAT	GGATCCCCTGCAGGCTACGCATCTCCTCTTCCCTTC	202
5R		
YSCAT 5	ATGCCTGCACCAAAACTCACGGAGAAATCTGCCTCTCCA	203
KO F	CTGTGCGGTATTCACACCG	
YSCAT 5	CTACGCATCTCCTCTTCCCTCTTCTTCTTCCCTCT	204
KO R	AGATTGTACTGAGAGTCAC	
YSCAT	GGATCCGGCGGCCGCACAATGTCTGCTCCGCTGCCGATCA	205
6F	TAACGCC	
YSCAT	GGATCCCCTGCAGGTCAATTCTTCTTCTGTGTTCTCTT	206
6R	TCTG	
YSCAT 6	ATGTCTGCTCCGCTGCCGATCATAACGCTGCCAACCTA	207
KO F	CTGTGCGGTATTCACACCG	
YSCAT 6	TCATTCTTCTTCTGTGTTCTCTTCTGTCTTACCAAGC	208
KO R	AGATTGTACTGAGAGTCAC	
YSCAT	GGATCCGGCGGCCGCACAATGCTGCATAAAAAATAGCTCA	209
7F	TAAAGTTCG	
YSCAT	GGATCCCCTGCAGGTCAAAAATAAAACAATAAAGTTAT	210
7R	AAACTAAC	
YSCAT 7	ATGCTGCATAAAAAATAGCTCATAAAGTTCGAAAAGTCG	211
KO F	CTGTGCGGTATTCACACCG	
YSCAT 7	TCAAAAATAAAACAATAAAGTTATAAACTAACCAAATT	212
KO R	AGATTGTACTGAGAGTCAC	
YSCAT	GGATCCGGCGGCCGCACAATGAGTGTGATAGGTAGGTTCTT	213
8F	G	
YSCAT	GGATCCCCTGCAGGTTAATGCATCTTTTACAGATGAAC	214
8R	C	
YSCAT 8	ATGAGTGTGATAGGTAGGTTCTGTATTACTTGAGGTCCG	215
KO F	CTGTGCGGTATTCACACCG	
YSCAT 8	TTAATGCATCTTTTACAGATGAACCTCGTTATGGGTA	216
KO R	AGATTGTACTGAGAGTCAC	

The entire coding regions for each of the acyltransferase sequences were amplified using the respective primers listed in the Table 3 above, cloned into the vector pCR2.1Topo (Invitrogen) or pZero (Invitrogen), and labeled as pCGN8558 (ATAT1), pCGN8564

(ATAT2), pCGB8565 (ATAT3), pCGN8566 (ATAT4), pCGN8918 (ATAT6), pCGN8913 (ATAT7), pCGN8904 (ATAT8), pCGN9970 (ATAT9), pCGN9940 (ATAT10), pCGN8567 (ATAT11), pCGN8632 (ATLPAAT1), pCGN9901 (YSCAT1 also referred to as gi2132299), pCGN9902 (YSCAT2, also referred to as gi1078509), pCGN9903 (YSCAT3, also referred to as gi2132939), pCGN9904 (YSCAT4, also referred to as gi2133031), pCGN9905 (YSCAT5, also referred to as gi320748), pCGN9906 (YSCAT6, also referred to as gi549627), pCGN9907 (YSCAT7, also referred to as gi586485), and pCGN9908 (YSCAT8, also referred to as gi464422). The nucleic acid sequences for the respective yeast acyltransferase are provided YSCAT1 (SEQ ID NO:225), YSCAT2 (SEQ ID NO:226), YSCAT3 (SEQ ID NO:227), YSCAT4 (SEQ ID NO:228), YSCAT5 (SEQ ID NO:229), YSCAT6 (SEQ ID NO:230), YSCAT7 (SEQ ID NO:231), and YSCAT8 (SEQ ID NO:232).

7A. Baculovirus Expression Constructs

Constructs are prepared to direct the expression of the *Arabidopsis* ATAT sequences in cultured insect cells. The entire coding regions of ATAT1, 2, 3, 4, 6, 7, 8, 9, 10, and 11 are cloned into the vector pFastBac1 (Gibco-BRL, Gaithersburg, MD) digested with *NotI* and 5 *PstI*. The respective coding sequences were cloned as *NotI/Sse8387I* fragments. Double stranded DNA sequence was obtained to verify that no errors were introduced by PCR amplification. The resulting plasmid were designated pCGN9723 (ATAT1), pCGN9724 (ATAT2), pCGN9725 (ATAT3), pCGN9726 (ATAT4), pCGN9727 (ATAT5), pCGN9728 (ATAT7), pCGN9729 (ATAT8), pCGN9991 (ATAT9) pCGN9730 (ATAT10), pCGN9731 10 (ATAT11).

7B. Plant Expression Construct Preparation

A plasmid containing the napin cassette derived from pCGN3223 (described in USPN 5,639,790, the entirety of which is incorporated herein by reference) was modified to make it 15 more useful for cloning large DNA fragments containing multiple restriction sites, and to allow the cloning of multiple napin fusion genes into plant binary transformation vectors. An adapter comprised of the self annealed oligonucleotide of sequence CGCGATTAAATGGCGGCCCTGCAGGCGGCCCTGCAGGGCGGCCATTAA (SEQ ID NO:233) AT was ligated into the cloning vector pBC SK+ (Stratagene) after 20 digestion with the restriction endonuclease BssHII to construct vector pCGN7765. Plamids pCGN3223 and pCGN7765 were digested with *NotI* and ligated together. The resultant vector, pCGN7770, contains the pCGN7765 backbone with the napin seed specific expression cassette from pCGN3223.

The cloning cassette, pCGN7787, essentially the same regulatory elements as 25 pCGN7770, with the exception of the napin regulatory regions of pCGN7770 have been replaced with the double CAMV 35S promoter and the tml polyadenylation and transcriptional termination region.

A binary vector for plant transformation, pCGN5139, was constructed from 30 pCGN1558 (McBride and Summerfelt, (1990) Plant Molecular Biology, 14:269-276). The polylinker of pCGN1558 was replaced as a HindIII/Asp718 fragment with a polylinker containing unique restriction endonuclease sites, *AscI*, *PacI*, *XbaI*, *SwaI*, *BamHI*, and *NotI*. The Asp718 and HindIII restriction endonuclease sites are retained in pCGN5139.

A series of turbo binary vectors are constructed to allow for the rapid cloning of DNA sequences into binary vectors containing transcriptional initiation regions (promoters) and transcriptional termination regions.

The plasmid pCGN8618 was constructed by ligating oligonucleotides 5'-

5 TCGAGGATCCGGCCGCAAGCTCCTGCAGG-3') (SEQ ID NO:234) and 5'-
TCGACCTGCAGGAAGCTTGCAGGCCGGATCC-3') (SEQ ID NO:235) into SalI/XhoI-digested pCGN7770. A fragment containing the napin promoter, polylinker and napin 3' region was excised from pCGN8618 by digestion with Asp718I; the fragment was blunt-ended by filling in the 5' overhangs with Klenow fragment then ligated into pCGN5139 that had been digested with Asp718I and HindIII and blunt-ended by filling in the 5' overhangs with Klenow fragment. A plasmid containing the insert oriented so that the napin promoter was closest to the blunted Asp718I site of pCGN5139 and the napin 3' was closest to the blunted HindIII site was subjected to sequence analysis to confirm both the insert orientation 10 and the integrity of cloning junctions. The resulting plasmid was designated pCGN8622.

15 The plasmid pCGN8619 was constructed by ligating oligonucleotides 5'-

TCGACCTGCAGGAAGCTTGCAGGCCGGATCC -3') (SEQ ID NO:236) and 5'-
TCGAGGATCCGGCCGCAAGCTCCTGCAGG-3') (SEQ ID NO:237) into SalI/XhoI-digested pCGN7770. A fragment containing the napin promoter, polylinker and napin 3' region was removed from pCGN8619 by digestion with Asp718I; the fragment was blunt-ended by filling in the 5' overhangs with Klenow fragment then ligated into pCGN5139 that had been digested with Asp718I and HindIII and blunt-ended by filling in the 5' overhangs with Klenow fragment. A plasmid containing the insert oriented so that the napin promoter was closest to the blunted Asp718I site of pCGN5139 and the napin 3' was closest to the blunted HindIII site was subjected to sequence analysis to confirm both the insert orientation 20 and the integrity of cloning junctions. The resulting plasmid was designated pCGN8623.

The plasmid pCGN8620 was constructed by ligating oligonucleotides 5'-

TCGAGGATCCGGCCGCAAGCTCCTGCAGGAGCT -3') (SEQ ID NO:238) and 5'-
CCTGCAGGAAGCTTGCAGGCCGGATCC-3') (SEQ ID NO:239) into SalI/SacI-digested pCGN7787. A fragment containing the d35S promoter, polylinker and tml 3' region 30 was removed from pCGN8620 by complete digestion with Asp718I and partial digestion with NotI. The fragment was blunt-ended by filling in the 5' overhangs with Klenow fragment then ligated into pCGN5139 that had been digested with Asp718I and HindIII and blunt-ended by filling in the 5' overhangs with Klenow fragment. A plasmid containing the insert

oriented so that the d35S promoter was closest to the blunted Asp718I site of pCGN5139 and the tml 3' was closest to the blunted HindIII site was subjected to sequence analysis to confirm both the insert orientation and the integrity of cloning junctions. The resulting plasmid was designated pCGN8624.

5 The plasmid pCGN8621 was constructed by ligating oligonucleotides 5'-
TCGACCTGCAGGAAGCTTGCAGGCCGGATCCAGCT -3') (SEQ ID NO:240) and 5'-
GGATCCGGCCGCAAGCTTCCTGCAGG-3') (SEQ ID NO:241) into SalI/SacI-
digested pCGN7787. A fragment containing the d35S promoter, polylinker and tml 3' region
was removed from pCGN8621 by complete digestion with Asp718I and partial digestion with
10 NotI. The fragment was blunt-ended by filling in the 5' overhangs with Klenow fragment
then ligated into pCGN5139 that had been digested with Asp718I and HindIII and blunt-
ended by filling in the 5' overhangs with Klenow fragment. A plasmid containing the insert
oriented so that the d35S promoter was closest to the blunted Asp718I site of pCGN5139 and
the tml 3' was closest to the blunted HindIII site was subjected to sequence analysis to
15 confirm both the insert orientation and the integrity of cloning junctions. The resulting
plasmid was designated pCGN8625.

 The coding regions of the various acyltransferase sequences were cloned as
NotI/Sse8387I fragments into pCGN8622, pCGN8623, pCGN8624, and pCGN8625, for
expression in sense or antisense orientations from a tissue preferential promoter, napin, or the
20 35S promoter. Fragments which were cloned into the pCGN8622 vector created the
constructs pCGN8901 (ATAT1), pCGN8571 (ATAT2), pCGN8909 (ATAT3), pCGN8596
(ATAT4), pCGN8919 (ATAT6), pCGN8914 (ATAT7), pCGN8905 (ATAT8), pCGN9973
(ATAT9), pCGN9942 (ATAT10), pCGN8575 (ATAT11), and pCGN8633 (ATLPAAT1) for
the sense expression of the respective coding sequences from the napin promoter. Fragments
25 which were cloned into the pCGN8623 vector created the constructs pCGN8900 (ATAT1),
pCGN8572 (ATAT2), pCGN8910 (ATAT3), pCGN8597 (ATAT4), pCGN8920 (ATAT6),
pCGN8915 (ATAT7), pCGN8906 (ATAT8), pCGN9972 (ATAT9), pCGN9943 (ATAT10),
pCGN8576 (ATAT11), and pCGN8634 (ATLPAAT1) for the antisense expression of the
respective coding sequences from the napin promoter. Fragments which were cloned into the
30 pCGN8624 vector created the constructs pCGN8903 (ATAT1), pCGN8573 (ATAT2),
pCGN8911 (ATAT3), pCGN8598 (ATAT4), pCGN8921 (ATAT6), pCGN8916 (ATAT7),
pCGN8907 (ATAT8), pCGN9971 (ATAT9), pCGN9944 (ATAT10), pCGN8577 (ATAT11),
and pCGN8635 (ATLPAAT1) for the sense expression of the respective coding sequences

from the 35S promoter. Fragments which were cloned into the pCGN8625 vector created the constructs pCGN8902 (ATAT1) and pCGN9974 (ATAT9) for the antisense expression of the respective coding sequences from the 35S promoter.

In addition, the yeast acyltransferase coding sequences were cloned into the vector pCGN8624 creating the constructs pCGN9926 (YSCAT1), pCGN9927 (YSCAT2), pCGN9928 (YSCAT3), pCGN9929 (YSCAT4), pCGN9930 (YSCAT5), pCGN9931 (YSCAT6), pCGN9932 (YSCAT7), and pCGN9933 (YSCAT8). These constructs allow for the sense expression of the respective acyltransferase coding sequences from the 35S promoter in plant cells.

10

Example 8: Plant Transformation

A variety of methods have been developed to insert a DNA sequence of interest into the genome of a plant host to obtain the transcription or transcription and translation of the sequence to effect phenotypic changes.

Transgenic *Brassica* plants are obtained by *Agrobacterium*-mediated transformation as described by Radke *et al.* (*Theor. Appl. Genet.* (1988) 75:685-694; *Plant Cell Reports* (1992) 11:499-505). Transgenic *Arabidopsis thaliana* plants may be obtained by 20 *Agrobacterium*-mediated transformation as described by Valverkens *et al.*, (*Proc. Nat. Acad. Sci.* (1988) 85:5536-5540), or as described by Bent *et al.* ((1994), *Science* 265:1856-1860), or Bechtold *et al.* ((1993), *C.R.Acad.Sci, Life Sciences* 316:1194-1199) or Clough, *et al.* (1998) *Plant J.*, 16:735-43. Other plant species may be similarly transformed using related techniques.

25 Alternatively, microprojectile bombardment methods, such as described by Klein *et al.* (*Bio/Technology* 10:286-291) may also be used to obtain nuclear transformed plants.

The above results demonstrate that the nucleic acid sequences identified encode proteins which are related to protein sequences encoding acyltransferase proteins. Such 30 acyltransferase sequences find use in preparing expression constructs for plant transformations.

All publications and patent applications mentioned in this specification are indicative of the level of skill of those skilled in the art to which this invention pertains. All

publications and patent applications are herein incorporated by reference to the same extent as if each individual publication or patent application was specifically and individually indicated to be incorporated by reference.

Although the foregoing invention has been described in some detail by way of illustration and example for purposes of clarity of understanding, it will be obvious that certain changes and modifications may be practiced within the scope of the appended claim.

Claims

What is Claimed is:

1. An isolated DNA sequence encoding an enzyme of the class of acyltransferase-like

5 proteins,

wherein said enzyme includes the amino acid sequence of SEQ ID NO: 127
(VxNHxS) wherein the H is the conserved Histidine residue in the conserved peptide
sequence HXXXXD of said acyltransferase-like protein, x representing any amino acid.

10 2. An isolated DNA sequence encoding an enzyme of the class of acyltransferase-like
proteins,

wherein said enzyme includes the amino acid sequence of SEQ ID NO: 128
(VTYSxS) within about 30 amino acids downstream from the conserved amino acid sequence
HXXXXD of said acyltransferase-like protein, x representing any amino acid.

15

3. An isolated DNA sequence encoding an enzyme of the class of acyltransferase-like
proteins,

wherein said enzyme includes the amino acid sequence of SEQ ID NO: 129
(VxLTRxR) within about 60 amino acids downstream from the conserved amino acid
sequence HXXXXD of said acyltransferase-like protein, x representing any amino acid.

20

4. An isolated DNA sequence encoding an enzyme of the class of acyltransferase-like
proteins,

wherein said enzyme includes the amino acid sequence of SEQ ID NO: 132
(LxxGDLV) within about 20 amino acids upstream of the conserved amino acid sequence
PEG of said acyltransferase-like protein, x representing any amino acid.

25

5. An isolated DNA sequence encoding an enzyme of the class of acyltransferase-like
proteins,

30

wherein said enzyme includes the amino acid sequence of SEQ ID NO: 130 (CPEGT)
containing the conserved amino acid sequence PEG of said acyltransferase-like protein.

6. An isolated DNA sequence encoding an enzyme of the class of acyltransferase-like proteins,

wherein said enzyme includes the amino acid sequence of SEQ ID NO: 133 (FxxGAF) within about 20 amino acids downstream from the conserved amino acid sequence PEG of said acyltransferase-like protein, x representing any amino acid.

7. An isolated DNA sequence encoding an enzyme of the class of acyltransferase-like proteins,

wherein said enzyme includes the amino acid sequence of SEQ ID NO: 131 (IVPVA) within about 40 amino acids downstream from the conserved amino acid sequence PEG of said acyltransferase-like protein.

8. An isolated DNA sequence encoding an enzyme of the class of acyltransferase-like proteins,

wherein said enzyme includes the amino acid sequence of SEQ ID NO: 134 (VANxxQ) within about 110 amino acids downstream from the conserved amino acid sequence PEG of said acyltransferase-like protein, x representing any amino acid.

20 9. A DNA sequence encoding an enzyme of the class of acyltransferase-like proteins, said DNA sequence obtainable by the steps comprising:

(a) using the profile of Figure 1 to search a nucleic acid sequence database;
(b) obtaining a probability score for nucleic acid sequences in said sequence database using the Smith-Waterman algorithm; and

25 (c) selecting a nucleic acid sequence having a probability score of less than about 1.

10. The DNA encoding sequence according to Claim 9, wherein said DNA sequence is an encoding sequence.

30 11. The DNA encoding sequence according to Claim 9, wherein said DNA sequence is an EST.

12. The DNA encoding sequence according to any one of Claims 1 to 11, wherein said acyltransferase-like protein is from a plant.

13. A construct comprising a DNA sequence of any one of Claims 1 to 11 linked to a
5 heterologous transcriptional and translational initiation region functional in a host cell.

14. The construct according to Claim 13 wherein said host cell is a plant cell.

15. A plant cell comprising a DNA construct according to Claim 13.

10 16. A plant comprising a cell according to Claim 15.

17. The DNA encoding sequence of any one of 1 to 11 wherein said acyltransferase-
15 like protein is from *Arabidopsis thaliana*.

18. The DNA encoding sequence of any one of 1 to 11 wherein said acyltransferase-
like protein is from corn.

20 19. The DNA encoding sequence of Claim 18 wherein said sequence comprises and
EST selected from the group consisting of SEQ ID NO: 86 through SEQ ID NO: 126.

20 . The DNA encoding sequence of any one of 1 to 11 wherein said acyltransferase-
like protein is from soybean.

25 21. The DNA encoding sequence of Claim 20 wherein said sequence comprises and
EST selected from the group consisting of SEQ ID NO: 24 through SEQ ID NO: 85.

30 22 . The DNA encoding sequence of any one of Claims 2, 3, 4, 5, 7 and 8 wherein
said acyltransferase-like protein is selected from the group consisting of SEQ ID NO: 1, SEQ
ID NO: 10, SEQ ID NO: 12, SEQ ID NO: 14 and SEQ ID NO: 16.

23 . The DNA encoding sequence of either of Claim 1 and Claim 6 wherein said acyltransferase-like protein is selected from the group consisting of SEQ ID NO: 3, SEQ ID NO: 5, SEQ ID NO: 7 and SEQ ID NO: 18.

Con	S	I	G	T	N	K	F	W	P	E	D	E	G	H	L	M	N	P	R	V	W	X	Y	Z	Gap	Len
!10	K	0	-1	-4	-1	2	-2	-3	-2	-1	-3	-2	-1	-2	-2	-1	0	-1	-1	-2	-3	-4	-5	-6	-5	1
!20	K	2	-2	-3	-4	-1	-4	-4	-3	-3	-3	-4	-1	-4	-2	-3	-1	0	-1	-2	-3	-4	-5	-6	-5	1
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Figure 3/5

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Figure 5/5

ATAT1	PLSB_ECOLI	160	IVP VAM NCK QGM FNGT	Figure 2
ATAT9	PLSB_MOUSE	160	IVP VAM NCK QGM FNGT	2/3
ATAT7	ATPAAT1	150	IVP VAM NCK QGM FNGT	
ATAT8	Jojoba_AT	150	IVP VAM NCK QGM FNGT	
ATAT6	Maize_AT	150	IVP VAM NCK QGM FNGT	
PLSB_ECOLI	ATAT11	140	IVP VAM NCK QGM FNGT	
PLSB_MOUSE	PLSC_COCO	140	IVP VAM NCK QGM FNGT	
ATPAAT1	PLSC_LM	140	IVP VAM NCK QGM FNGT	
Jojoba_AT	PLSC_ECOLI	140	IVP VAM NCK QGM FNGT	
Maize_AT	PLSC_YEAST	140	IVP VAM NCK QGM FNGT	
ATAT11	ATAT2	130	IVP VAM NCK QGM FNGT	
PLSC_COCO	ATAT3	130	IVP VAM NCK QGM FNGT	
PLSC_LM	ATAT4	130	IVP VAM NCK QGM FNGT	
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ATAT7	T A R G W K G L D P I	
ATAT8	T A S G L K A L D P L	
ATAT6	T A S G L K A F D P I	
PLSB_MOUSE	M E V G	
ATLPAAT1	V S N M R S F V P A I	180
Jojoba_AT	V S H M R S F V P A I	
Maize_AT	V S I M R D F V P A I	
ATAT11	L Q E L S C S L D A V Y	
PLSC_COCO	S L - R V R P A P I	
PLSC_LIM	T F - R V R P V P I	
PLSC_ECOLI	L N - R L H N G L V	
PLSC_YEAST	Y G - V F N R G C M	
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ATAT3	W D T I S G A R H I L	
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	Q L M T S W A V V C E V W Y L E P Q T I R P	
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	N P F L L K E L R G A	
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	Q L G K P K K N E S L W	
	S S S T C R G V P D	
	- Q M L R G	
	- F K G Q P S	200
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	- T K D K I G	
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	- F K G Q P S	210
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	- F K G Q P S	
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	- L K G Q S S	
	- Y G I E P S	
	- E E E K I N	
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	- G K D Q V R	
	- T K D K I G	
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ATAT8	D G K L K F E V A N N V Q S D I G K A L D F E	
ATAT11	N G K V N F E V A N H V Q H E I G N	
PLSB_MOUSE	L S K L R N L G Q G Y V R V D F A Q P F S L K E	230
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Maize_AT	V - - - - I H V R M K R H A M S E M P K S D E D V S K	
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PLSC_COCO	H - - - - Y V E M I H A L Y V D H L P E S Q K P L V	240
PLSC_LIM	D - - - - Y V K M I H D I Y V R N L P A S Q K P L G	
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	Figure 2	
	3/3	

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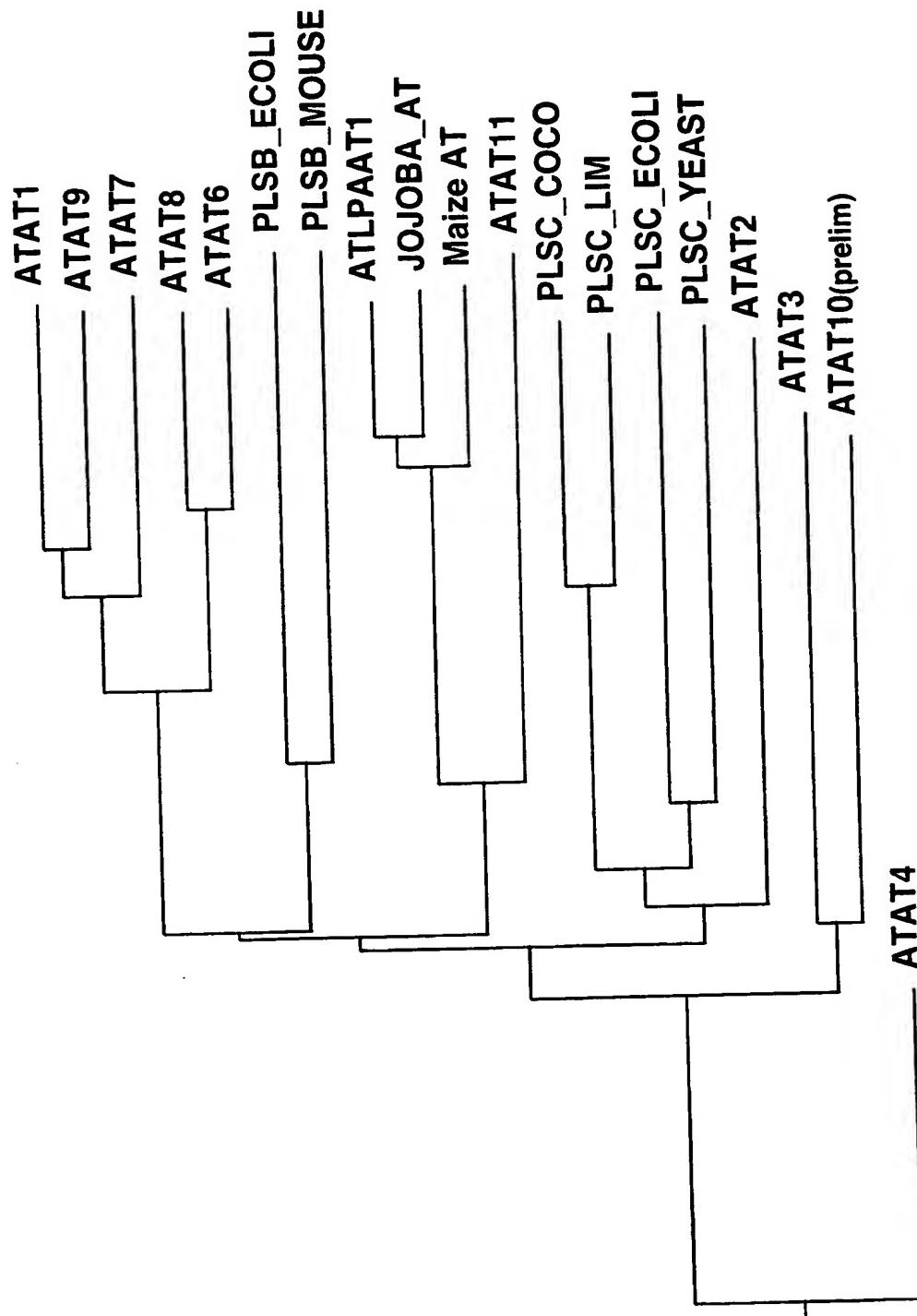


Figure 3 1/2

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3	40.2	35.8	44.8	44.8	12.9	14.4	14.9	13.4	11.3	12.9	12.4	12.4	12.9	11.9	13.9	13.4	17.1	14.4	3		
4	49.7	50.0	50.3		67.2	10.8	13.3	11.8	11.8	10.8	16.4	11.8	11.8	12.8	13.3	12.3	17.4	15.1	12.8	4	
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6	85.6	86.3	85.6	86.2	86.1		28.5	12.6	12.1	11.6	9.7	13.9	14.3	14.8	11.8	17.6	13.5	12.3	10.6	6	
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8	82.9	78.4	81.2	81.2	83.1	83.6	85.1		82.3	78.0	31.6	12.4	12.8	13.3	15.8	13.9	12.2	16.4	14.4	8	
9	83.5	77.8	81.8	85.9	84.6	85.6	87.1	18.2		77.5	32.1	11.9	14.3	13.3	16.3	15.5	12.4	15.1	12.0	9	
10	83.5	82.4	84.1	87.6	85.1	84.4	87.1	22.5		22.5	30.6	13.9	16.7	12.8	16.3	14.4	12.9	16.4	12.0	10	
11	84.1	81.4	83.1	85.1	90.3	87.4	65.4	65.0		67.5	14.4	14.8	11.8	12.8	13.4	14.6	15.8	12.9	11		
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18	78.5	82.5	81.7	81.8	88.7	87.1	79.1	80.5		78.9	82.8	81.8	78.1	76.1	78.1	79.3	64.8		18.5	18	
19	84.7	84.8	84.7	85.5	86.5	83.6	87.4	86.5		87.6	91.0	85.0	83.1	85.7	81.8	83.7	79.4	74.1		19	
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Figure 3 2/2

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Emig, Robin A
Ruezinsky, Diane
Van Eenennaam, Alison

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SEQUENCE LISTING

<110> Lassner, Michael W
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Van Eenennaam, Alison

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 Lys Leu Ala Val Ala Leu Val Thr Leu Val Pro Leu Arg Phe Leu Leu
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 Leu Phe Ser Ala Pro Tyr Arg Gly Pro Glu Glu Glu Asp Glu Gly
 100 105 110
 Gly Val Val Phe Gln Glu Asp Tyr Ala His Met Glu Gly Trp Lys Arg
 115 120 125
 Thr Val Ile Val Arg Ser Gly Arg Phe Leu Ser Arg Val Leu Leu Phe
 130 135 140
 Val Phe Gly Phe Tyr Trp Ile His Glu Ser Cys Pro Asp Arg Asp Ser
 145 150 155 160
 Asp Met Asp Ser Asn Pro Lys Thr Thr Ser Thr Glu Ile Asn Gln Lys
 165 170 175
 Gly Glu Ala Ala Thr Glu Glu Pro Glu Arg Pro Gly Ala Ile Val Ser
 180 185 190
 Asn His Val Ser Tyr Leu Asp Ile Leu Tyr His Met Ser Ala Ser Phe
 195 200 205
 Pro Ser Phe Val Ala Lys Arg Ser Val Gly Lys Leu Pro Leu Val Gly
 210 215 220
 Leu Ile Ser Lys Cys Leu Gly Cys Val Tyr Val Gln Arg Glu Ala Lys
 225 230 235 240
 Ser Pro Asp Phe Lys Gly Val Ser Gly Thr Val Asn Glu Arg Val Arg

245

250

255

Glu Ala His Ser Asn Lys Ser Ala Pro Thr Ile Met Leu Phe Pro Glu
 260 265 270

Gly Thr Thr Asn Gly Asp Tyr Leu Leu Thr Phe Lys Thr Gly Ala
 275 280 285

Phe Leu Ala Gly Thr Pro Val Leu Pro Val Ile Leu Lys Tyr Pro Tyr
 290 295 300

Glu Arg Phe Ser Val Ala Trp Asp Thr Ile Ser Gly Ala Arg His Ile
 305 310 315 320

Leu Phe Leu Leu Cys Gln Val Val Asn His Leu Glu Val Ile Arg Leu
 325 330 335

Pro Val Tyr Tyr Pro Ser Gln Glu Glu Lys Asp Asp Pro Lys Leu Tyr
 340 345 350

Ala Ser Asn Val Arg Lys Leu Met Ala Thr Glu Gly Asn Leu Ile Leu
 355 360 365

Ser Glu Leu Gly Leu Ser Asp Lys Arg Ile Tyr His Ala Thr Leu Asn
 370 375 380

Gly Asn Leu Ser Gln Thr Arg Asp Phe His Gln Lys Glu Glu
 385 390 395

<210> 7

<211> 1131

<212> DNA

<213> Arabidopsis sp.

<400> 7

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<211> 376

<212> PRT

<213> Arabidopsis sp.

<400> 8

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Asn Glu Pro Arg Gly Lys Leu Ser Leu Arg Asp Leu Leu Asp Ile Ser
35 40 45

Pro Thr Leu Thr Glu Ala Ala Gly Ala Ile Val Asp Asp Ser Phe Thr
50 55 60

Arg Cys Phe Lys Ser Asn Pro Pro Glu Pro Trp Asn Trp Asn Ile Tyr
65 70 75 80

Leu Phe Pro Leu Tyr Cys Phe Gly Val Val Val Arg Tyr Cys Ile Leu
85 90 95

Phe Pro Leu Arg Cys Phe Thr Leu Ala Phe Gly Trp Ile Ile Phe Leu
100 105 110

Ser Leu Phe Ile Pro Val Asn Ala Leu Leu Lys Gln Asp Arg Leu
115 120 125

Arg Lys Lys Ile Glu Arg Val Leu Val Glu Met Ile Cys Ser Phe Phe
130 135 140

Val Ala Ser Trp Thr Gly Val Val Lys Tyr His Gly Pro Arg Pro Ser
145 150 155 160

Ile Arg Pro Lys Gln Val Tyr Val Ala Asn His Thr Ser Met Ile Asp
165 170 175

Phe Ile Val Leu Glu Gln Met Thr Ala Phe Ala Val Ile Met Gln Lys
180 185 190

His Pro Gly Trp Val Gly Leu Leu Gln Ser Thr Ile Leu Glu Ser Val
195 200 205

Gly Cys Ile Trp Phe Asn Arg Ser Glu Ala Lys Asp Arg Glu Ile Val
210 215 220

Ala Lys Lys Leu Arg Asp His Val Gln Gly Ala Asp Ser Asn Pro Leu
225 230 235 240

Leu Ile Phe Pro Glu Gly Thr Cys Val Asn Asn Asn Tyr Thr Val Met
245 250 255

Phe Lys Lys Gly Ala Phe Glu Leu Asp Cys Thr Val Cys Pro Ile Ala
260 265 270

Ile Lys Tyr Asn Lys Ile Phe Val Asp Ala Phe Trp Asn Ser Arg Lys
275 280 285

Gln Ser Phe Thr Met His Leu Leu Gln Leu Met Thr Ser Trp Ala Val
290 295 300

Val Cys Glu Val Trp Tyr Leu Glu Pro Gln Thr Ile Arg Pro Gly Glu
305 310 315 320

Thr Gly Ile Glu Phe Ala Glu Arg Val Arg Asp Met Ile Ser Leu Arg
325 330 335

Ala Gly Leu Lys Lys Val Pro Trp Asp Gly Tyr Leu Lys Tyr Ser Arg
340 345 350

Pro Ser Pro Lys His Ser Glu Arg Lys Gln Gln Ser Phe Ala Glu Ser
355 360 365

Ile Leu Ala Arg Leu Glu Glu Lys
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<211> 965

<212> DNA
 <213> Arabidopsis sp.

<400> 9

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gccatggctc	gtcaattcca	tggaaatcat	caaattccta	aggttcttga	tcagactcta	300
cgaccatc	tccgttcttgc	tctatcttca	gagggaaacga	agaaacaggg	gaagaagata	360
aagaaaatgc	ggttcgcggaa	taatgtgaaa	gatacggaaag	gtaacgggga	agagtaccgg	420
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gatttagtt	ttgttaatct	ttctttgtt	tttcggtat	attagatttt	ttcttgaaa	660
tttcagatat	tgttagactt	gtagttgggt	ggtcttctt	ttctccctt	ttgtgtctca	720
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gatgtaaata	attgacatgt	aagttagtcat	tagaaatttg	aaaaggcaaa	tgaaagaata	840
taaatttgt	aaaacatagt	gtgcctattt	tacatataaa	ctctctttt	ttgggatata	900
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<211> 1593

<212> DNA

<213> Arabidopsis sp.

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gtgataaggt	cactttcct	cttagttctt	tatccattta	taagcttgc	gagctacgaa	300
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<211> 530

<212> PRT

<213> Arabidopsis sp.

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Lys Tyr Gln Lys Cys Pro Ser His Gly Leu His Gln Tyr Gln Asp Leu
35 40 45
Ser Asn His Thr Leu Ile Phe Asn Val Glu Gly Ala Leu Leu Lys Ser
50 55 60
Asn Ser Leu Phe Pro Tyr Phe Met Val Val Ala Phe Glu Ala Gly Gly
65 70 75 80
Val Ile Arg Ser Leu Phe Leu Leu Val Leu Tyr Pro Phe Ile Ser Leu
85 90 95
Met Ser Tyr Glu Met Gly Leu Lys Thr Met Val Met Leu Ser Phe Phe
100 105 110
Gly Val Lys Lys Glu Ser Phe Arg Val Gly Lys Ser Val Leu Pro Lys
115 120 125
Tyr Phe Leu Glu Asp Val Gly Leu Glu Met Phe Gln Val Leu Lys Arg
130 135 140
Gly Gly Lys Arg Val Ala Val Ser Asp Leu Pro Gln Val Met Ile Asp
145 150 155 160
Val Phe Leu Arg Asp Tyr Leu Glu Ile Glu Val Val Val Gly Arg Asp
165 170 175
Met Lys Met Val Gly Gly Tyr Tyr Leu Gly Ile Val Glu Asp Lys Lys
180 185 190
Asn Leu Glu Ile Ala Phe Asp Lys Val Val Gln Glu Glu Arg Leu Gly
195 200 205
Ser Gly Arg Arg Leu Ile Gly Ile Thr Ser Phe Asn Ser Pro Ser His
210 215 220
Arg Ser Leu Phe Ser Gln Phe Cys Gln Glu Ile Tyr Phe Val Arg Asn
225 230 235 240
Ser Asp Lys Ser Trp Gln Thr Leu Pro Gln Asp Gln Tyr Pro Lys
245 250 255
Pro Leu Ile Phe His Asp Gly Arg Leu Ala Val Lys Pro Thr Pro Leu
260 265 270
Asn Thr Leu Val Leu Phe Met Trp Ala Pro Phe Ala Ala Val Leu Ala
275 280 285
Ala Ala Arg Leu Val Phe Gly Leu Asn Leu Pro Tyr Ser Leu Ala Asn
290 295 300
Pro Phe Leu Ala Phe Ser Gly Ile His Leu Thr Leu Thr Val Asn Asn
305 310 315 320
His Asn Asp Leu Ile Ser Ala Asp Arg Lys Arg Gly Cys Leu Phe Val
325 330 335
Cys Asn His Arg Thr Leu Leu Asp Pro Leu Tyr Ile Ser Tyr Ala Leu
340 345 350
Arg Lys Lys Asn Met Lys Ala Val Thr Tyr Ser Leu Ser Arg Leu Ser

355

360

365

Glu Leu Leu Ala Pro Ile Lys Thr Val Arg Leu Thr Arg Asp Arg Val
 370 375 380

Lys Asp Gly Gln Ala Met Glu Lys Leu Leu Ser Gln Gly Asp Leu Val
 385 390 395 400

Val Cys Pro Glu Gly Thr Thr Cys Arg Glu Pro Tyr Leu Leu Arg Phe
 405 410 415

Ser Pro Leu Phe Ser Glu Val Cys Asp Val Ile Val Pro Val Ala Ile
 420 425 430

Asp Ser His Val Thr Phe Phe Tyr Gly Thr Thr Ala Ser Gly Leu Lys
 435 440 445

Ala Phe Asp Pro Ile Phe Phe Leu Leu Asn Pro Phe Pro Ser Tyr Thr
 450 455 460

Val Lys Leu Leu Asp Pro Val Ser Gly Ser Ser Ser Ser Thr Cys Arg
 465 470 475 480

Gly Val Pro Asp Asn Gly Lys Val Asn Phe Glu Val Ala Asn His Val
 485 490 495

Gln His Glu Ile Gly Asn Ala Leu Gly Phe Glu Cys Thr Asn Leu Thr
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Arg Arg Asp Lys Tyr Leu Ile Leu Ala Gly Asn Asn Gly Val Val Lys
 515 520 525

Lys Lys
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<211> 1509
<212> DNA
<213> Arabidopsis sp.

<400> 12

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<212> PRT
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Leu Val Ala Phe Glu Ala Ala Gly Leu Ile Arg Phe Ala Ile Leu Leu
35 40 45
Phe Leu Trp Pro Val Ile Thr Leu Leu Asp Val Phe Ser Tyr Lys Asn
50 55 60
Ala Ala Leu Lys Leu Lys Ile Phe Val Ala Thr Val Gly Leu Arg Glu
65 70 75 80
Pro Glu Ile Glu Ser Val Ala Arg Ala Val Leu Pro Lys Phe Tyr Met
85 90 95
Asp Asp Val Ser Met Asp Thr Trp Arg Val Phe Ser Ser Cys Lys Lys
100 105 110
Arg Val Val Val Thr Arg Met Pro Arg Val Met Val Glu Arg Phe Ala
115 120 125
Lys Glu His Leu Arg Ala Asp Glu Val Ile Gly Thr Glu Leu Ile Val
130 135 140
Asn Arg Phe Gly Phe Val Thr Gly Leu Ile Arg Glu Thr Asp Val Asp
145 150 155 160
Gln Ser Ala Leu Asn Arg Val Ala Asn Leu Phe Val Gly Arg Arg Pro
165 170 175
Gln Leu Gly Leu Gly Lys Pro Ala Leu Thr Ala Ser Thr Asn Phe Leu
180 185 190
Ser Leu Cys Glu Glu His Ile His Ala Pro Ile Pro Glu Asn Tyr Asn
195 200 205
His Gly Asp Gln Gln Leu Gln Leu Arg Pro Leu Pro Val Ile Phe His
210 215 220
Asp Gly Arg Leu Val Lys Arg Pro Thr Pro Ala Thr Ala Leu Ile Ile
225 230 235 240
Leu Leu Trp Ile Pro Phe Gly Ile Ile Leu Ala Val Ile Arg Ile Phe
245 250 255
Leu Gly Ala Val Leu Pro Leu Trp Ala Thr Pro Tyr Val Ser Gln Ile
260 265 270
Phe Gly Gly His Ile Ile Val Lys Gly Lys Pro Pro Gln Pro Pro Ala
275 280 285
Ala Gly Lys Ser Gly Val Leu Phe Val Cys Thr His Arg Thr Leu Met

290

295

300

Asp Pro Val Val Leu Ser Tyr Val Leu Gly Arg Ser Ile Pro Ala Val
 305 310 315 320

Thr Tyr Ser Ile Ser Arg Leu Ser Glu Ile Leu Ser Pro Ile Pro Thr
 325 330 335

Val Arg Leu Thr Arg Ile Arg Asp Val Asp Ala Ala Lys Ile Lys Gln
 340 345 350

Gln Leu Ser Lys Gly Asp Leu Val Val Cys Pro Glu Gly Thr Thr Cys
 355 360 365

Arg Glu Pro Phe Leu Leu Arg Phe Ser Ala Leu Phe Ala Glu Leu Thr
 370 375 380

Asp Arg Ile Val Pro Val Ala Met Asn Tyr Arg Val Gly Phe Phe His
 385 390 395 400

Ala Thr Thr Ala Arg Gly Trp Lys Gly Leu Asp Pro Ile Phe Phe Phe
 405 410 415

Met Asn Pro Arg Pro Val Tyr Glu Ile Thr Phe Leu Asn Gln Leu Pro
 420 425 430

Met Glu Ala Thr Cys Ser Ser Gly Lys Ser Pro His Asp Val Ala Asn
 435 440 445

Tyr Val Gln Arg Ile Leu Ala Ala Thr Leu Gly Phe Glu Cys Thr Asn
 450 455 460

Phe Thr Arg Lys Asp Lys Tyr Arg Val Leu Ala Gly Asn Asp Gly Thr
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Phe Glu Pro Cys Leu His
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<211> 1563

<212> DNA

<213> Arabidopsis sp.

<400> 14

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 1563

<210> 15
 <211> 520
 <212> PRT
 <213> Arabidopsis sp.

<400> 15
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 Gly Pro Ser Ser Leu Leu Gln Ser Asp Leu Ser Arg His Thr Leu Ile
 35 40 45
 Phe Asn Val Glu Gly Ala Leu Leu Lys Ser Asp Ser Leu Phe Pro Tyr
 50 55 60
 Phe Met Leu Val Ala Phe Glu Ala Gly Gly Val Ile Arg Ser Phe Leu
 65 70 75 80
 Leu Phe Ile Leu Tyr Pro Leu Ile Ser Leu Met Ser His Glu Met Gly
 85 90 95
 Val Lys Val Met Val Met Val Ser Phe Phe Gly Ile Lys Lys Glu Gly
 100 105 110
 Phe Arg Ala Gly Arg Ala Val Leu Pro Lys Tyr Phe Leu Glu Asp Val
 115 120 125
 Gly Leu Glu Ile Phe Glu Val Leu Lys Arg Gly Gly Lys Lys Ile Gly
 130 135 140
 Val Ser Asp Asp Leu Pro Gln Val Met Ile Glu Gly Phe Leu Arg Asp
 145 150 155 160
 Tyr Leu Glu Ile Asp Val Val Gly Arg Glu Met Lys Val Val Gly
 165 170 175
 Gly Tyr Tyr Leu Gly Ile Met Glu Asp Lys Thr Lys His Asp Leu Val
 180 185 190
 Phe Asp Glu Leu Val Arg Lys Glu Arg Leu Asn Thr Gly Arg Val Ile
 195 200 205
 Gly Ile Thr Ser Phe Asn Thr Ser Leu His Arg Tyr Leu Phe Ser Gln
 210 215 220
 Phe Cys Gln Glu Ile Tyr Phe Val Lys Lys Ser Asp Lys Arg Ser Trp
 225 230 235 240
 Gln Thr Leu Pro Arg Ser Gln Tyr Pro Lys Pro Leu Ile Phe His Asp
 245 250 255

Gly Arg Leu Ala Ile Lys Pro Thr Leu Met Asn Thr Leu Val Leu Phe
 260 265 270

Met Trp Gly Pro Phe Ala Ala Ala Ala Ala Ala Arg Leu Phe Val
 275 280 285

Ser Leu Cys Ile Pro Tyr Ser Leu Ser Ile Pro Ile Leu Ala Phe Ser
 290 295 300

Gly Cys Arg Leu Thr Val Thr Asn Asp Tyr Val Ser Ser Gln Lys Gln
 305 310 315 320

Lys Pro Ser Gln Arg Lys Gly Cys Leu Phe Val Cys Asn His Arg Thr
 325 330 335

Leu Leu Asp Pro Leu Tyr Val Ala Phe Ala Leu Arg Lys Lys Asn Ile
 340 345 350

Lys Thr Val Thr Tyr Ser Leu Ser Arg Val Ser Glu Ile Leu Ala Pro
 355 360 365

Ile Lys Thr Val Arg Leu Thr Arg Asp Arg Val Ser Asp Gly Gln Ala
 370 375 380

Met Glu Lys Leu Leu Thr Glu Gly Asp Leu Val Val Cys Pro Glu Gly
 385 390 395 400

Thr Thr Cys Arg Glu Pro Tyr Leu Leu Arg Phe Ser Pro Leu Phe Thr
 405 410 415

Glu Val Ser Asp Val Ile Val Pro Val Ala Val Thr Val His Val Thr
 420 425 430

Phe Phe Tyr Gly Thr Thr Ala Ser Gly Leu Lys Ala Leu Asp Pro Leu
 435 440 445

Phe Phe Leu Leu Asp Pro Tyr Pro Thr Tyr Thr Ile Gln Phe Leu Asp
 450 455 460

Pro Val Ser Gly Ala Thr Cys Gln Asp Pro Asp Gly Lys Leu Lys Phe
 465 470 475 480

Glu Val Ala Asn Asn Val Gln Ser Asp Ile Gly Lys Ala Leu Asp Phe
 485 490 495

Glu Cys Thr Ser Leu Thr Arg Lys Asp Lys Tyr Leu Ile Leu Ala Gly
 500 505 510

Asn Asn Gly Val Val Lys Lys Asn
 515 520

<210> 16
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gtgaaaaacat tccttaggggt tgataaagtt cttggAACAG agcttagaggt ctccaaatcg 480
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agcaagacgg accacgactt catgtccatc tgcaaggaag gttacatggc gccacgtacg 660

aaatgcgaac cattaccaag aaacaaactc ttaagcccc taatattcca cgagggcaga 720
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 ttgcgtctct ctatcatccg cgtctacacg aatattccgt taccggaaacg tatcgcccg 840
 tacaactaca agcttactgg catcaagcta gtcgtcaacg gccaccctcc tccgcccgg 900
 aaacctggcc agccaggcca tctttggtc tgcaaccacc gcaccgttct cgatccgtg 960
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 1140
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 1200
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 1260
 aagcttcttg atccttactt tgcgttcatg aacccgaggc cgacgtatga gatcacgttc
 1320
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 1380
 aattacatac agagggttt gggaggaacc ttaggtttg agtgcaccaa tttcacaaga
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 aaggataagt acgcaatgct tgctggtaact gacggtaggg ttccggtgaa gaaggagaag
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 1506

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 <212> PRT
 <213> *Arabidopsis* sp.

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 Thr Leu Leu Ile Ser Arg Ser Ala Phe Pro Tyr Tyr Phe Leu Val Ala
 35 40 45
 Leu Glu Ala Gly Ser Leu Leu Arg Ala Leu Ile Leu Leu Val Ser Val
 50 55 60
 Pro Phe Val Tyr Leu Thr Tyr Leu Thr Ile Ser Glu Thr Leu Ala Ile
 65 70 75 80
 Asn Val Phe Val Phe Ile Thr Phe Ala Gly Leu Lys Ile Arg Asp Val
 85 90 95
 Glu Leu Val Val Arg Ser Val Leu Pro Arg Phe Tyr Ala Glu Asp Val
 100 105 110
 Arg Pro Asp Thr Trp Arg Ile Phe Asn Thr Phe Gly Lys Arg Tyr Ile
 115 120 125
 Ile Thr Ala Ser Pro Arg Ile Met Val Glu Pro Phe Val Lys Thr Phe
 130 135 140
 Leu Gly Val Asp Lys Val Leu Gly Thr Glu Leu Glu Val Ser Lys Ser
 145 150 155 160
 Gly Arg Ala Thr Gly Phe Thr Arg Lys Pro Gly Ile Leu Val Gly Gln
 165 170 175
 Tyr Lys Arg Asp Val Val Leu Arg Glu Phe Gly Gly Leu Ala Ser Asp
 180 185 190
 Leu Pro Asp Leu Gly Leu Gly Asp Ser Lys Thr Asp His Asp Phe Met
 195 200 205

Ser Ile Cys Lys Glu Gly Tyr Met Val Pro Arg Thr Lys Cys Glu Pro
 210 215 220
 Leu Pro Arg Asn Lys Leu Leu Ser Pro Ile Ile Phe His Glu Gly Arg
 225 230 235 240
 Leu Val Gln Arg Pro Thr Pro Leu Val Ala Leu Leu Thr Phe Leu Trp
 245 250 255
 Leu Pro Val Gly Phe Val Leu Ser Ile Ile Arg Val Tyr Thr Asn Ile
 260 265 270
 Pro Leu Pro Glu Arg Ile Ala Arg Tyr Asn Tyr Lys Leu Thr Gly Ile
 275 280 285
 Lys Leu Val Val Asn Gly His Pro Pro Pro Pro Lys Pro Gly Gln
 290 295 300
 Pro Gly His Leu Leu Val Cys Asn His Arg Thr Val Leu Asp Pro Val
 305 310 315 320
 Val Thr Ala Val Ala Leu Gly Arg Lys Ile Ser Cys Val Thr Tyr Ser
 325 330 335
 Ile Ser Lys Phe Ser Glu Leu Ile Ser Pro Ile Lys Ala Val Ala Leu
 340 345 350
 Thr Arg Gln Arg Glu Lys Asp Ala Ala Asn Ile Lys Arg Leu Leu Glu
 355 360 365
 Glu Gly Asp Leu Val Ile Cys Pro Glu Gly Thr Thr Cys Arg Glu Pro
 370 375 380
 Phe Leu Leu Arg Phe Ser Ala Leu Phe Ala Glu Leu Thr Asp Arg Ile
 385 390 395 400
 Val Pro Val Ala Ile Asn Thr Lys Gln Ser Met Phe Asn Gly Thr Thr
 405 410 415
 Thr Arg Gly Tyr Lys Leu Leu Asp Pro Tyr Phe Ala Phe Met Asn Pro
 420 425 430
 Arg Pro Thr Tyr Glu Ile Thr Phe Leu Lys Gln Ile Pro Ala Glu Leu
 435 440 445
 Thr Cys Lys Gly Gly Lys Ser Pro Ile Glu Val Ala Asn Tyr Ile Gln
 450 455 460
 Arg Val Leu Gly Gly Thr Leu Gly Phe Glu Cys Thr Asn Phe Thr Arg
 465 470 475 480
 Lys Asp Lys Tyr Ala Met Leu Ala Gly Thr Asp Gly Arg Val Pro Val
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 500

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 <212> DNA
 <213> Arabidopsis sp.

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 ctcagcgagt cagagccccc ggttctcggt ccgacgacgg tggatccatt ccggaacaat 240
 acacctggag tttagcggatt gtacgaagcg attaagctcg tgatgtct tccgattgct 300

ctgattagac ttgttctctt tgctgcttagc ttagctgttg gttacttggc tacaaaattg 360
 gcacttgcgt gctggaaaga taaagagaac cctatgcctc tttggagatg cagaatcatg 420
 tggattactc ggatctgtac cagatgtatc ctcttcctt ttggctatca gtggataaga 480
 aggaaaggga aacctgcgtc gagagagatt gctccgattt ttgtatcaa tcatgttct 540
 tatattgaac caatcttcta cttctatgaa ttatcaccga ccattgttc atcgagatca 600
 catgattcac ttccatttgt tggacttatt atcaggcga tgcaggtat atatgtaat 660
 agatctcac agacatcaag gaagaatgtc gtgcataaaa taaagagaaa agcttctgc 720
 gatagatttc ctcgtctgtc ttattccccca gaaggaacca cgactaatgg gaaagttctt 780
 atttccctcc aactcgggtc tttcatccct gttacccta tcaacacgtt agtagtccgg 840
 tatccccatg tacatttga tcaatcctgg gaaaaatatct cttgttgc gctcatgtt 900
 agaatgttca ctcagttca caatttcatg gaggttgaat atcttcctgt aatctatccc 960
 agtggaaaagc aaaagcagaa tgctgtgcgt ctctcacaga agactagtca tgcaattgca
 1020
 acatcttga atgtcgtcca aacatccat tctttgcgg acttgatgtc actcaacaaa
 1080
 gcaactgagt taaagctgga gaacccctca aattacatgg ttgaaatggc aagagttgag
 1140
 tcgcatttcc atgtaagcag ctttagggca acgcgatttt tggatacatt tgttccatg
 1200
 attccggact cgagtggacg tggtaggcta catgacttcc ttcgggtct taaactgaaa
 1260
 ccttgcctc tttctaaaag gatattttag ttcatcgatg tggagaaggc cgatcaatc
 1320
 acttcaaac agttcttgc tgcctcgggc cacgttga cacagccgct ttttaagcaa
 1380
 acatgcgagc tagcctttc ccattgcgt gcagatggag atggctatat tacaattcaa
 1440
 gaactcggag aagctctcaa aaacacaatc ccaaacttga acaaggacga gattcgagga
 1500
 atgtaccatt tgcttagacga cgaccaagat caaagaatca gccaaaatga cttgttgc
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 1620

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 <211> 539
 <212> PRT
 <213> *Arabidopsis* sp.

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Ser Gly Leu Asn Leu Leu Pro Ala Val Val Asp Pro Arg Val Ser Arg			
35	40	45	
Gly Phe Glu Phe Asp His Leu Asn Pro Tyr Gly Phe Leu Ser Glu Ser			
50	55	60	
Glu Pro Pro Val Leu Gly Pro Thr Thr Val Asp Pro Phe Arg Asn Asn			
65	70	75	80
Thr Pro Gly Val Ser Gly Leu Tyr Glu Ala Ile Lys Leu Val Ile Cys			
85	90	95	
Leu Pro Ile Ala Leu Ile Arg Leu Val Leu Phe Ala Ala Ser Leu Ala			
100	105	110	
Val Gly Tyr Leu Ala Thr Lys Leu Ala Leu Ala Gly Trp Lys Asp Lys			
115	120	125	
Glu Asn Pro Met Pro Leu Trp Arg Cys Arg Ile Met Trp Ile Thr Arg			
130	135	140	
Ile Cys Thr Arg Cys Ile Leu Phe Ser Phe Gly Tyr Gln Trp Ile Arg			
145	150	155	160

Arg Lys Gly Lys Pro Ala Arg Arg Glu Ile Ala Pro Ile Val Val Ser
 165 170 175
 Asn His Val Ser Tyr Ile Glu Pro Ile Phe Tyr Phe Tyr Glu Leu Ser
 180 185 190
 Pro Thr Ile Val Ala Ser Glu Ser His Asp Ser Leu Pro Phe Val Gly
 195 200 205
 Thr Ile Ile Arg Ala Met Gln Val Ile Tyr Val Asn Arg Phe Ser Gln
 210 215 220
 Thr Ser Arg Lys Asn Ala Val His Glu Ile Lys Arg Lys Ala Ser Cys
 225 230 235 240
 Asp Arg Phe Pro Arg Leu Leu Leu Phe Pro Glu Gly Thr Thr Asn
 245 250 255
 Gly Lys Val Leu Ile Ser Phe Gln Leu Gly Ala Phe Ile Pro Gly Tyr
 260 265 270
 Pro Ile Gln Pro Val Val Val Arg Tyr Pro His Val His Phe Asp Gln
 275 280 285
 Ser Trp Gly Asn Ile Ser Leu Leu Thr Leu Met Phe Arg Met Phe Thr
 290 295 300
 Gln Phe His Asn Phe Met Glu Val Glu Tyr Leu Pro Val Ile Tyr Pro
 305 310 315 320
 Ser Glu Lys Gln Lys Gln Asn Ala Val Arg Leu Ser Gln Lys Thr Ser
 325 330 335
 His Ala Ile Ala Thr Ser Leu Asn Val Val Gln Thr Ser His Ser Phe
 340 345 350
 Ala Asp Leu Met Leu Leu Asn Lys Ala Thr Glu Leu Lys Leu Glu Asn
 355 360 365
 Pro Ser Asn Tyr Met Val Glu Met Ala Arg Val Glu Ser Leu Phe His
 370 375 380
 Val Ser Ser Leu Glu Ala Thr Arg Phe Leu Asp Thr Phe Val Ser Met
 385 390 395 400
 Ile Pro Asp Ser Ser Gly Arg Val Arg Leu His Asp Phe Leu Arg Gly
 405 410 415
 Leu Lys Leu Lys Pro Cys Pro Leu Ser Lys Arg Ile Phe Glu Phe Ile
 420 425 430
 Asp Val Glu Lys Val Gly Ser Ile Thr Phe Lys Gln Phe Leu Phe Ala
 435 440 445
 Ser Gly His Val Leu Thr Gln Pro Leu Phe Lys Gln Thr Cys Glu Leu
 450 455 460
 Ala Phe Ser His Cys Asp Ala Asp Gly Asp Gly Tyr Ile Thr Ile Gln
 465 470 475 480
 Glu Leu Gly Glu Ala Leu Lys Asn Thr Ile Pro Asn Leu Asn Lys Asp
 485 490 495
 Glu Ile Arg Gly Met Tyr His Leu Leu Asp Asp Asp Gln Asp Gln Arg
 500 505 510
 Ile Ser Gln Asn Asp Leu Leu Ser Cys Leu Arg Arg Asn Pro Leu Leu
 515 520 525
 Ile Ala Ile Phe Ala Pro Asp Leu Ala Pro Thr

530

535

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<211> 1128
<212> DNA
<213> Arabidopsis sp.

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ttatcagctg tagtgtttag gctttcagc attcgatata gccgttaatg ttttccttc 180
ttctttggct cgtggctcgc cttgtggcct ttcccttttga agaagattaa caaaaccaa 240
gttatcttct ctggtgataa ggttcccttgc gaggatcgag tattgctcat tgcaaacac 300
cgaacagaag ttgattggat gtacttctgg gatcttgcac tgcgttaaagg ccagattggg 360
aatatcaat atgtcttaa gagtagttt atgaaattac ctcttttgg ttggcggtt 420
caccttttgc agtttatcc ttttgagagg agatggaaag tcgtatgaaagc aaacttgaga 480
cagatagttt ctagttttaa ggatccccga gacgctttat ggcttgcctt tttcccccgg 540
ggcacagatt acacagaggc taaatgcca aggagtaaga aatttgcgc tgaaaatggc 600
cttccgatac tgaacaacgt gctgtttccc aggacaaaag gtttgcgc tgcgttgc 660
gaactgagtt gctcaacttga cgcaatgtt gatgtgacca tcggttataa aaccgcgtc 720
ccatctttct tagacaacgt ttatggatt gagccatcag aagttcacat ccacatccgt 780
cgtatcaacc tgacccaaat cccaaatcaa gaaaaggaca tcaatgcctt gttaatgaa 840
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gaaggaaacag agaaaagagtt caacacaaaag aagtacctca taaactgtttt ggcagtgatt 960
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1128

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<211> 375
<212> PRT
<213> Arabidopsis sp.

<400> 21
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20 25 30
Met Met Leu Ile Phe Trp Gly Phe Leu Ser Ala Val Val Leu Arg Leu
35 40 45
Phe Ser Ile Arg Tyr Ser Arg Lys Cys Val Ser Phe Phe Phe Gly Ser
50 55 60
Trp Leu Ala Leu Trp Pro Phe Leu Phe Glu Lys Ile Asn Lys Thr Lys
65 70 75 80
Val Ile Phe Ser Gly Asp Lys Val Pro Cys Glu Asp Arg Val Leu Leu
85 90 95
Ile Ala Asn His Arg Thr Glu Val Asp Trp Met Tyr Phe Trp Asp Leu
100 105 110
Ala Leu Arg Lys Gly Gln Ile Gly Asn Ile Lys Tyr Val Leu Lys Ser
115 120 125
Ser Leu Met Lys Leu Pro Leu Phe Gly Trp Ala Phe His Leu Phe Glu
130 135 140
Phe Ile Pro Val Glu Arg Arg Trp Glu Val Asp Glu Ala Asn Leu Arg
145 150 155 160
Gln Ile Val Ser Ser Phe Lys Asp Pro Arg Asp Ala Leu Trp Leu Ala
165 170 175

Leu Phe Pro Glu Gly Thr Asp Tyr Thr Glu Ala Lys Cys Gln Arg Ser
 180 185 190
 Lys Lys Phe Ala Ala Glu Asn Gly Leu Pro Ile Leu Asn Asn Val Leu
 195 200 205
 Leu Pro Arg Thr Lys Gly Phe Val Ser Cys Leu Gln Glu Leu Ser Cys
 210 215 220
 Ser Leu Asp Ala Val Tyr Asp Val Thr Ile Gly Tyr Lys Thr Arg Cys
 225 230 235 240
 Pro Ser Phe Leu Asp Asn Val Tyr Gly Ile Glu Pro Ser Glu Val His
 245 250 255
 Ile His Ile Arg Arg Ile Asn Leu Thr Gln Ile Pro Asn Gln Glu Lys
 260 265 270
 Asp Ile Asn Ala Trp Leu Met Asn Thr Phe Gln Leu Lys Asp Gln Leu
 275 280 285
 Leu Asn Asp Phe Tyr Ser Asn Gly His Phe Pro Asn Glu Gly Thr Glu
 290 295 300
 Lys Glu Phe Asn Thr Lys Lys Tyr Leu Ile Asn Cys Leu Ala Val Ile
 305 310 315 320
 Ala Phe Thr Thr Ile Cys Thr His Leu Thr Phe Phe Ser Ser Met Ile
 325 330 335
 Trp Phe Arg Ile Tyr Val Ser Leu Ala Cys Val Tyr Leu Thr Ser Ala
 340 345 350
 Thr His Phe Asn Leu Arg Ser Val Pro Leu Val Glu Thr Ala Lys Asn
 355 360 365
 Ser Leu Lys Leu Val Asn Lys
 370 375

<210> 22
 <211> 1170
 <212> DNA
 <213> Arabidopsis sp.

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 tacagaaaaaa ttaaccgggt ggttgcagaa accttgggt tggagcttgg atggatagtt 180
 gactgggtgg ctggagttaa gatccaagt tttgctgata atgagacctt caatcgaatg 240
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 tggattctgg ctcagcggtc aggttgcctt ggaagcgcat tagctgtaat gaagaagtct 360
 tccaaattcc ttccagtcat aggctggtca atgtggttct cggagtatct ctttctggaa 420
 agaaaattggg ccaaggatga aaggactcta aagtcaaggc ttcagcgctt gagcgacttc 480
 cctcgacctt tctggtagc ccttttgtg gagggaaactc gctttacaga agccaaactt 540
 aaagccgcac aagagtatgc agcctcttctt gaattgccta tccctcgaaa tgtgttggatt 600
 cctcgacca aaggtttgcgt gtcaagctgtt agtaatatgc gtcattttgt cccagcaatt 660
 tatgatatga cagtgaactat tccaaaaaac tctccaccac ccacgatgtc aagactattc 720
 aaagacaac cttcagttggt gcatgttacat atcaagtgtc actcgatgaa agacttacat 780
 gaatcgatg acgcaatgtc acatgggtgc agagatcgtt ttgtggctaa ggatgctctg 840
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 aagtccctac actgggcaca actctttctt tcataggaaag gtatcacat atcggcgctt
 1020
 ggtcttaggtt tcatcactct ctgtatgcag atcctgatac gctcgtctca gtcagagcgt
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 1140

tcccaaacag aaacggagaa ggagaagtaa
1170

<210> 23
<211> 389
<212> PRT
<213> Arabidopsis sp.

<400> 23
Met Val Ile Ala Ala Ala Val Ile Val Pro Leu Gly Leu Leu Phe Phe
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20 25 30
Ile Arg Pro Leu Ser Lys Asn Thr Tyr Arg Lys Ile Asn Arg Val Val
35 40 45
Ala Glu Thr Leu Trp Leu Glu Leu Val Trp Ile Val Asp Trp Trp Ala
50 55 60
Gly Val Lys Ile Gln Val Phe Ala Asp Asn Glu Thr Phe Asn Arg Met
65 70 75 80
Gly Lys Glu His Ala Leu Val Val Cys Asn His Arg Ser Asp Ile Asp
85 90 95
Trp Leu Val Gly Trp Ile Leu Ala Gln Arg Ser Gly Cys Leu Gly Ser
100 105 110
Ala Leu Ala Val Met Lys Lys Ser Ser Lys Phe Leu Pro Val Ile Gly
115 120 125
Trp Ser Met Trp Phe Ser Glu Tyr Leu Phe Leu Glu Arg Asn Trp Ala
130 135 140
Lys Asp Glu Ser Thr Leu Lys Ser Gly Leu Gln Arg Leu Ser Asp Phe
145 150 155 160
Pro Arg Pro Phe Trp Leu Ala Leu Phe Val Glu Gly Thr Arg Phe Thr
165 170 175
Glu Ala Lys Leu Lys Ala Ala Gln Glu Tyr Ala Ala Ser Ser Glu Leu
180 185 190
Pro Ile Pro Arg Asn Val Leu Ile Pro Arg Thr Lys Gly Phe Val Ser
195 200 205
Ala Val Ser Asn Met Arg Ser Phe Val Pro Ala Ile Tyr Asp Met Thr
210 215 220
Val Thr Ile Pro Lys Thr Ser Pro Pro Pro Thr Met Leu Arg Leu Phe
225 230 235 240
Lys Gly Gln Pro Ser Val Val His Val His Ile Lys Cys His Ser Met
245 250 255
Lys Asp Leu Pro Glu Ser Asp Asp Ala Ile Ala Gln Trp Cys Arg Asp
260 265 270
Gln Phe Val Ala Lys Asp Ala Leu Leu Asp Lys His Ile Ala Ala Asp
275 280 285
Thr Phe Pro Gly Gln Gln Glu Gln Asn Ile Gly Arg Pro Ile Lys Ser
290 295 300
Leu Ala Val Val Leu Ser Trp Ala Cys Val Leu Thr Leu Gly Ala Ile
305 310 315 320
Lys Phe Leu His Trp Ala Gln Leu Phe Ser Ser Trp Lys Gly Ile Thr

325

330

335

Ile Ser Ala Leu Gly Leu Gly Ile Ile Thr Leu Cys Met Gln Ile Leu
340 345 350

Ile Arg Ser Ser Gln Ser Glu Arg Ser Thr Pro Ala Lys Val Val Pro
355 360 365

Ala Lys Pro Lys Asp Asn His His Pro Glu Ser Ser Ser Gln Thr Glu
370 375 380

Thr Glu Lys Glu Lys
385

<210> 24
<211> 269
<212> DNA
<213> Glycine max

<400> 24
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ataagggtct acttcaacct ccctctccca gaacncattg tccgctacac ctacgagatg 120
ctcgccatca acctcgtcat ccgcggccac cgccctcctc cgccctcccc cggcacccccc 180
ggcaacctct acgtctgc aa ccacccgacc gctctcgacc ccatcgtcat cgccattgcc 240
ctcgcccgca aggtctccctg cgtcaccta 269

<210> 25
<211> 242
<212> DNA
<213> Glycine max

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tcccagaacg cattgtccgc tacacctacg agatgctcgg catcaacctc gtcatccg 180
gccacccgccc tcctccgcct tccccggca ccccccggcaa cctctacgctc tgcaaccacc 240
gc 242

<210> 26
<211> 272
<212> DNA
<213> Glycine max

<400> 26
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catcataactc tccatnctta agggtctacc ttaacatccc ttgcctgaa agaattgctt 120
ggtataaacta taagctatta ggaatcagag ttattgtgaa ggttacccctt ccaccacccc 180
caaagaagggt tcaaagtgtt gtccattttt tttgttaacca ccgcacagtt ttagaccctg 240
tggttactgc agttgcactt ggaagaaaaa tt 272

<210> 27
<211> 218
<212> DNA
<213> Glycine max

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cgtctgaaga gcagaatgtat cttccacgac gggcgttcg tgagaggcc agaccaatg 120
aatgcctca tcaccttcac atggctccct ttgggttgc tcctctccat cataagggtc 180
tacttcaacc tccctctccc agaacgcata gtcgcata 218

<210> 28
<211> 270
<212> DNA
<213> Glycine max

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aagttctggg acccttaact tacttcttac atgaacccta ggcctgtgtc cgaggttacc 120

ttaccttgat accttgcgg aggagatgtc ggttaaggct ggggggaagt cgtccattga 180
ggtggccaac cacgtggcaag aagggtgctgg gggatgtgtt agggttttag tgcaccgggt 240
tgacttaggaa ggataagtat atgttgttgg 270

<210> 29
<211> 252
<212> DNA
<213> Glycine max

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ctgccaattg gcatcatact ctccatctta agggtctacc ttaacatccc ttgcctgaa 120
agaatttggt gtacaactac aagctcttag gaatcagagt tattgtgaag ggtacccttc 180
caccccccccc aaaaagggtt caaatgggt tctatggtt tctaaccacc gcacagtatt 240
agaccctgtt gt 252
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<210> 30
<211> 272
<212> DNA
<213> Glycine max

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<400> 30
ctgggactgc cttaaacgat gcatggatct tatcaagaaa ggagcctctg ttttttctt 60
tccagaggga acacgcgata aagatggaag actaggcaca ttcaagaagg gtgcatttcag 120
tgtgtctca aagacaaaatg caccagtatg accaattacc ttatggaa ctggtaaat 180
catggctca gggaaaggagg gaatagtgaa cataggttct gtgaaaagtgg ttatacataa 240
acctatgtt gggaaaggatc ctgacatgtt at 272
```

<210> 31
<211> 239
<212> DNA
<213> Glycine max

```
<400> 31
cgggaaatcaa ggtcatcaga cttcaagggt gtttcagctg ttgtcaactga cagaattcga 60
gaagctcata agaatggatc tgctccatta atatgttat ttccagaagg tacaaccacca 120
aatggagagt tcctccattc attcaagact ggtggtttt tgccaaaggc accggtactt 180
ccttgatatac tacqatatac ttaccqaqaa tttaqcctq ccttggatc catatctgg 239
```

<210> 32
<211> 242
<212> DNA
<213> Glycine max

```
<400> 32
gaacggcaac ggcaaacagcg ttgcgcgatga ccgtcctctg ctgaagccgg agcctccgg 60
cttccgcgcga cagcatcgcc gatatggaga agaagttcgc cgcttacgtc cgccgcgtacg 120
tgtacggcac catgggacgc ggcgagttgc ctccccaaagga gaagctcttg ctcggtttcg 180
cgttggtcac tcttcctcccc attcgagtcg ttctcgccgt caccatattt ctcttttattt 240
ac                                         242
```

<210> 33
<211> 248
<212> DNA
<213> Glycine max

```
<400> 33
ttcttcttct ctcactctct aaaaccctaa ctctatacat ggaagggaaa nctcaaatct 60
natgactaat taattaatcc atcgatcaag catggagtcc gaactcaaag acctcaattc 120
gaagccgcgg aacggcaacg gcaacagcgt tcgcgatgac cgttctctgc tgaagccgga 180
gcctccggtc tccggcgaca gcatgccgat tatggagaag aagttcgccg cttacgtccg 240
ccggcgaca 248
```

<210> 34
<211> 217
<212> DNA
<213> Glycine max

<400> 34
aaaacccctaa ttctatacat ggaaggggaaa tctcaaattct aatgactaat taatttaatcc 60

atcgatcaag catggagtcc gaactcaaag acctcaattc gaagccgccc aacggcaacg 120
gcaacagcgt tcgcgatgac cgtccctctgc tgaagccgga gcctccggtc tccggcaca 180
gcatcgccga tatggagaag aagttcgccg cttacgt 217

<210> 35
<211> 257
<212> DNA
<213> Glycine max

<400> 35
atctctgtct ctgcatttcc ctccctaaaa ccctaattct acatttggaa agggaaatctc 60
aaatctaattg actaattaat caatcaatcg tattaataat ccatcgatca agtatggagt 120
ccgaactcaa agacctcaat tcgaagccac ccaactgcaa cggcaacgccc aacagcgaaa 180
gcgacgaccg tcctctgtc aagccggagc ctccggcctc ctccgacagc atcgccgaga 240
tggagaagaa gttcgcc 257

<210> 36
<211> 284
<212> DNA
<213> Glycine max

<400> 36
cccgacccaa acagggtttt gtggccaatc atacttccat gattgatttc attatcttag 60
aacagatgac tgcatttgct ttattatgc agaagcatcc tggatgggtt ggattattgc 120
agagcaccat tntggaggt gtaggggtga tctgggtcaa ccgtacagag gcaaaggatc 180
gagaagttgt ggcaaggaaa ttgagggatc atgtccctggg agctaacaac aaccctcttc 240
ttatatttcc tgaaggaact tgtgtaaata atcactactc gtca 284

<210> 37
<211> 246
<212> DNA
<213> Glycine max

<400> 37
ggagatccgc ataagcaaat caatcatcct gttccttcct tatctctgtc tctgcatttc 60
cctccctaaa accctaattc tacatttggaa aaggaantct caaatctaatt gataattaat 120
caatcaatcg tattaataat ccatcgatca agtatggagt ccgaactcaa agacctcaat 180
tcgaagccac ccaactgcaa cggcaacgccc aacagcgaaa gcgacgaccg tcctctgctg 240
aagccg 246

<210> 38
<211> 278
<212> DNA
<213> Glycine max

<400> 38
gtttctatt gccacgttgt ggaagcgtaa cgaagatgaa tggcattggg aaactcaaatt 60
cgtcgagttc tgaattggac cttcacattt aagattacct accttctggaa tccagtggttc 120
aacaagaacg gcatggcaag ctccgactgt gtgatttgct agacatttct cctagtctat 180
ctgagggcagc acgtgccatt gtagatgata cattcacaag gtgcttcaag caaatcttc 240
agaaccttgg aacttggaaatg tttattttgtt tcctttgtt 278

<210> 39
<211> 312
<212> DNA
<213> Glycine max

<400> 39
ttaaccttgg cacatttcc tttgttcat caatgtgtgt tgtaaattgt ncatttcctt 60
cagaggtctt tggtaganat gatgtgcagt ttctgtgggt catctggac tgngngtgg 120
aagnatcatg gacccaggcc tagcaggaga ccaaagcagg tttttagtc caaccatact 180
tcatgattga tntcattatn tnagaacaga tgactgcttt tgcngrttn atgcagaagc 240
atcctggatg ggttggtaag cttacagnat gtcaacngtg tatnaaatat gntacacnnn 300
acttgcgtct tc 312

<210> 40
<211> 255
<212> DNA
<213> Glycine max

<400> 40
ggattattgn ngcanaatgca gtcatctgtt ctaagataat ganatcnatc atggaagtat 60
gattggncac anaaacctgt yttttgggtt gataacttagt cttggcccat ggtacttgac 120
naccaggc catgatgcaa canaganact gnacatcatc tccaccaaac ccctctgana 180
ganacgagaa ttgagcaatt tagagtacct tggtttgatg caagtcagta tattcaagtt 240
tctattcatc aaagg 255

<210> 41
<211> 291
<212> DNA
<213> Glycine max

<400> 41
caacctccca tgcaatcgct caccctctcc gtcacactgaa tctgtttctt attccctccg 60
tcgcgtaaca aggtgaatg gcattggaa actcaatcg tcgagttctg aattggacct 120
tcacattgaa gattacctgc cttctggatc cagtgttcaa caagaacggc atggcaagct 180
ccgcctgtgt gatttgcgt acatttctcc tagtctatct gaggcagcac gtgccattgt 240
agatgataca ttcacaaggc gcttcaagtc aaatcccca gaaccttggaa a 291

<210> 42
<211> 284
<212> DNA
<213> Glycine max

<400> 42
ctgcaaccta ccatgcaatt cctcacctga atccgttttc tattgccacg ttgttggaa 60
gtaacgaaga tgaatggcat tggaaactc aaatcgatc gttctgaatt ggacccatc 120
attgaagatt acctacccctc tggatccagt gttcaacaag aacggcatgg caagctccga 180
ctgtgtgatt tgcttagacat ttctccttagt ctatctgagg cagcacgtgc catgtagatg 240
atacatcaca aggtgctcaa gtcaaattctc cagaaccttg gaat 284

<210> 43
<211> 268
<212> DNA
<213> Glycine max

<400> 43
ctgaagtatt ctcgtcctag cccaaagcat agagaaaaggn agcaacagaa ctttgcttag 60
tcagtgcgtc ggcgatggga gaaaaagtga tgtgtacctt tatgtgggtg tggcttaat 120
tattcttagt aatgccattt cttcgacccc ttttttgct tttgtttgtt cattgctaac 180
tatttatttt taacactttt attaaagata tggcatatat ncacttcagt anacaaagtt 240
gtncaggtaa ttntttcc aaaaaaaaaa 268

<210> 44
<211> 241
<212> DNA
<213> Glycine max

<400> 44
gancaaattt gcccctccatc actttccttg ttagagttgg tttctgcnaac ctaccatgca 60
attccctcac ctgaatccgt ttcttattgc cacgtgtgg aacgctaacg aagatgaatg 120
gcattggaa actcaaatcg tcgagttctg aattggacct tcacattgaa gattacctac 180
cttctggatc cagtgttcaa caagaacggc atggcaagct ccgactgtgt gatttgcttag 240
a 241

<210> 45
<211> 247
<212> DNA
<213> Glycine max

<400> 45
ttaggatgtc tgagatccctt gccccaaatca aaacggtgcg gttactaga aaccgcgacg 60
aggatgcgaa aatgatgaaa aatttgcgtt ggcggggaa cttgggtgt tggcttgcag 120
ggaccacatg tagagaacctt tattttatgtt ggttcagccc tctgttctca gagatgtgcg 180
atgagatgtt ccccggtggc agttgattcc cagttatatg ttccacggaa ccactgctgg 240
tgganta 247

<210> 46
<211> 271
<212> DNA

<213> Glycine max

<400> 46

tgcagggggg cttgttagag ccatagtttt gggtcttcta taccctttt tttgtgtcg 60
aggaaaagag atggggttga agataatggt catggcatgc ttcttcggga tcaaagcatc 120
gagcttcaga gttggaaaggc ccgtttgcc cnaattctc tngaggacg ttngtgcaga 180
aatgtttgag gcactcaaaa aaggaggaa gacagtggga gtacccaatt taccacgt 240
gatggtgaa agttcttga gagagtattt g 271

<210> 47

<211> 242

<212> DNA

<213> Glycine max

<400> 47

ttcacagctg tcacggcgtn aacggaaaat ggcaacggcg agacgcagtt tccgcctat 60
caccgaatgc aacggAACGA cnccgtgcga ntctgtngnc gccgacctcg agggtacgct 120
cctcatctcc cgtnctcg tccgtactt catgctcg tcgcgtcaag ccggcagcnt 180
cctccggc ctcatgtc tcctctccct tccgttcgtc atnatgcct acctttcat 240
ct 242

<210> 48

<211> 244

<212> DNA

<213> Glycine max

<400> 48

acatattctt cagttagctc ccccaaccta tacacttcac caccacacca caaccctacc 60
ctctctctct gtcatggtca ttggaggagc cttccctcg ttcgacccaa tcaccaatg 120
tagacccaag accgctccaa ccagaccatc gcctcgacc tcgatggcac ctccttg 180
tccccggatg cttccctta ctacttcctc gtcgcctcg aagccggcag cgtttccga 240
gcct 244

<210> 49

<211> 230

<212> DNA

<213> Glycine max

<400> 49

caacattcca cctagctccc caatcacatc ttaccacac cataaacctt cttatattct 60
ctcttcattt tctcctctat tgcataatc atggggacct tccctcgct cgacccaatc 120
accacccaag accgggtccaa ccagaccgtg gcctccgacc ttgacggcac ctcctcg 180
tccccggagcg cttccctta ctacccctc gttgcctcg aagccggcag 230

<210> 50

<211> 265

<212> DNA

<213> Glycine max

<400> 50

ctggtaata atcctaagtt atggagtctg tgggtgtgtga gctagaaggc acgcttgtga 60
aggacaagga tgcgttctca tacttcatgt tggttgcgtt tgaagcttca gttttgggtc 120
gtttcgccctt gttgctaaca ctattgccc tgattcggtt cttgacatg gttggcatga 180
acgatgcac tctcaagcta ntatctcg tggctgtggc tgggttccca aagtccgaga 240
ttgaatcagt ggctaggc gttt 265

<210> 51

<211> 252

<212> DNA

<213> Glycine max

<400> 51

ctggtaata atcctaagtt atggagtctg tgggtgtgtga gctagaaggc acgcttgtga 60
aggacaagga tgcgttctca tacttcatgt tggttgcgtt tgaagcttca gttttgggtc 120
gtttcgccctt gttgctaaca ctattgccc tgattcggtt cttgacatg gttggcatga 180
acgatgcac tctcaagcta atgatctcg tggctgtggc tgggttccca agtccgagat 240
tgaatcagtgc 252

<210> 52

<211> 218

<212> DNA

<213> Glycine max

<400> 52

aactgcaact acaaacaat tcattcattc acagctgtca cgccgtgaac gaaaaatggc 60
aacggcgaga cgcagttac ccgcctatac accgaatgca acggaacgac accgtgcgag 120
tctgtggcg ccgacctcga cggtagctc ctcatntccc gtagctcggtt cccgtacttc 180
atgctcgctg ccgtcgaagc cggcagcctc ctccgcgg 218

<210> 53

<211> 262

<212> DNA

<213> Glycine max

<400> 53

ggtaaggac attgagatgg tcgnntcctc ggtgctgccc aagttctaca ccgaggacgt 60
gcncggcag agctggagag tcttaatcc ttccggaaagc gttacattgt cactgctagt 120
ctagggtat ggtggagcan ttgttaaga cgtttcttgg ggctgataag gtgcttggga 180
ctgagcttga ggccacgaaa tcggggaggt tcatgggtt gttaggagc ctggtgtgct 240
tggtgggag cacaagaaag tg 262

<210> 54

<211> 212

<212> DNA

<213> Glycine max

<400> 54

gcaactacaa caacattcat tcattcacag ctgtcacgcc gtgaacggaa aatggcaacg 60
gcgagacgca gtttccgccc tatcaccgaa tgcaacggaa cgacgccgtg cgagtctgtg 120
gccggcgacc tcgacggtac gctcctcatc tcccgtagn cgttcccgta cttcatgctc 180
gtngccgtcg aagccggcag ctcctccgc gg 212

<210> 55

<211> 273

<212> DNA

<213> Glycine max

<400> 55

catggtttc ttgagcttct ttggcctcag aaaggacaca ttcagaacag gatcagctgt 60
tctgcaaag ttcttcttag aagatgttgg attggaaggc ttgaggccg taatatgtt 120
tgagagaaaa gtggcatcta gtaagttgcc aagggtcatg gttaaaatt tcctcaagga 180
ctattnaggg ttgatgtg ttatagcaag agaattgaag tcctttagtg gttttttttt 240
gggagttttt gagagtaaga agccaattaa aat 273

<210> 56

<211> 257

<212> DNA

<213> Glycine max

<400> 56

ctctcaaaaa aggagggaaag acagtggag tcaccaatct accccatgtg atggggaaaa 60
gcttctttag agagtatttgc gacattgatt tcgttgggg cagggagctg aaagttttct 120
gtggatacta cgtaggatttgc atggatgaca caaaaactat gcatgcctt gagctggta 180
aagaaggaaa aggatgtcc gacatgtcg gaatcacaag gtttcgcaac atacgcgacc 240
atgtatgtttttt tttctcc 257

<210> 57

<211> 240

<212> DNA

<213> Glycine max

<400> 57

gaactaagtgc tgaaccacta ccaagaaaaca agcttttaag tccaatttatt tttcatgagg 60
gttagtttgc tcaaaggcca actcctctag ctgnnctttt gaccccttca tggctgccaa 120
ttggcatcat actctccatc ttaagggtct accttaacat cccttgcct gaaagaattt 180
cttggtacaa ctacaagtc ttaggaatca gagttattgtt gaaagggtacc cttccaccgc 240

<210> 58

<211> 254

<212> DNA

<212> DNA

<213> Glycine max

<400> 64

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ccgagaaccg gtctaaccaa accgtggcct cggacttggaa cggcacccctc ctgggtgtccc 60
ccagcgccatt tccttactat atgctggtcg ccatacgaa cggcagcttc ctccgtggcc 120
ttgtccctct tgcctccgtc ccttcgtgt attcacgtac atattccctc ccgagaccgc 180
ggccatcaag tccctgatct tcatacgccct cgcgggcctg aaggtaggg acgttgagat 240
ggtcgcgtgc tcgggtgcgtc ccaagttcta cgccgacata ttcttcgttgtt agctccccca 300
acctatacac ttcaccacca caccacaaacc ctaccctctc ttcttcgtcat ggtcattggaa 360
ggagccttcc ctcgttgcga cccaaatcacc aaatgttagca cccaaagacccg ctccaaaccag 420
accatcgccct cggacctcga tggcaccctc cttgtctccc ggagtgcctt cccctactac 480
ttcctcgatcg ccctcgaa cggcagcgcc ttccgagcccc tccttcgtt a 531
```

<210> 65

<211> 256

<212> DNA

<213> Glycine max

<400> 65

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acatattctt cagtttagctc ccccaaccta tacacttcac caccacacca caaccctacc 60
ctctctctct gtcatggta ttggaggagc cttccctcgtt ttcgacccaa tcacccaaatg 120
tagcacccaa gaccgctcca accagaccat cgcctcggac ctcgatggca ccctccctgt 180
ctcccgaggt gccttccct actacttcct cgtccctcga gaagccggca ggcgttccg 240
agccctcctt ctctta 256
```

<210> 66

<211> 260

<212> DNA

<213> Glycine max

<400> 66

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ccatccaaca tatttttcag ttagctcccc caacctatac acttcaccac cacaccacaa 60
cccttaccctc tctctctgtc atggtcattt gaggagccctt ccctcgttt gacccaaatca 120
ccaaatgttag caccctaaac cgtccaaacc agactatcgc ctcggacccctt gatggcacc 180
tccttgcgtc ccggagtgcc ttccctactt acttcctcgtt cgcctcga gccggcagcg 240
tcttccgagc cctccttctc 260
```

<210> 67

<211> 248

<212> DNA

<213> Glycine max

<400> 67

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caccaaccaa acctcactct ccctttctcc cctgaccctc tccctgccc ggtcatggga 60
gcctttggcc acttcgaacc ggtctccaaa tgcagcaccg agaaccggtc taaccaaacc 120
gtggcctcggt acttggacgg caccctctgt gtgtccccc ggcattttcc ttactacatg 180
ctgggcgccca tcgaagccgg cagcttccctc cgtggccttgc tcctcccttgc ctccgtccct 240
ttcgtgtt 248
```

<210> 68

<211> 283

<212> DNA

<213> Glycine max

<400> 68

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ttcttccca ccatcacacc aancaaaccc cactctnccctt ggcctatggtc atgnnnngcct 60
ttccgcact tcgaaccgggt ttccaaatgc agcaccggaaa accggttttaa ccaaaccgtg 120
gcctcggact tggacggcac cttccctgggt tcccttagcg cttttccctta ctacatgctc 180
gtgcctcatcg aagccggcag cttccctccgtt ggccttgcc tccttggatc cgtcccttcc 240
gtgtacttca cgtacatattt cttctccgag accggccca tca 283
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<210> 69

<211> 258

<212> DNA

<213> Glycine max

<400> 69

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ctcttcttcc ccaccatcnn accaaccaaa cctcactctc cctgaccatg gtcatgggag 60
ccttccgcca cttcgaaccg gttccaaat gcagcaccga aaaccgggtt aaccaaaccg 120
```

tggcctcgga cttggacggc accctcctgg tgcctccat cgccttcct tactacatgc 180
tcgtcgccat cgaagccggc agtctcctcc gtggccctgt ctccttgcga tccgtccctt 240
tcgtgtactt cacgtaca 258

<210> 70
<211> 256
<212> DNA
<213> Glycine max

<400> 70
tgcaactaca acaacattca ttcattcaca gctgtcacgc cgtgaacggaaaatggcaac 60
ggcgagacgc agtttccgc ctatcaccga atgcaacggaa acgacaccgt gcgagtctgt 120
ggccgccgac ctcgacggta cgctcctcat ctcccgttagc tcgttccctgt acttcatgtct 180
cgtcgccgac gaagccggca gcntcctccg cggcctcatc ctccctctng ccantccgtt 240
cgtcatcanc gcctac 256

<210> 71
<211> 259
<212> DNA
<213> Glycine max

<400> 71
cttccccacc atcacacccan ggcnaacctc antctccctt tctccacnaga ccctctccct 60
gccatngtca tggancctt tggccacttc gaaccggctt ccaaatgcag caccgagaac 120
cggnctaacc aaaccgtggc ctcggacttg gacggcaccc tcctgggtgtcc cnncagcga 180
tttccttact acatgctggc ngccatcgaa gccggcagct tcctccgtgg ctttgtccctc 240
cttgccctcg tccctttcg 259

<210> 72
<211> 249
<212> DNA
<213> Glycine max

<400> 72
ccaacatatt cttcagtttag ctcccccac ctatacactt caccaccaca ccacaaccct 60
accctctctc tctgtcatgg tcattggagg agcctccct cgtttcgacc caatcaccaa 120
atgttagcacc caagaccgct ccaaccagac catcgccctcg gacctcgatg gcaccctnct 180
tgtctcccg agtgccttcc cctactactt cctcgtcgcc ctgcgaagccg gcagcgtctt 240
ncgagccct 249

<210> 73
<211> 257
<212> DNA
<213> Glycine max

<400> 73
caacccttctt cttccccacc atcacaccaa ncaaacctca ctctccctt ctcccctgac 60
cctccctgt ccatggtcat gggagcctt ggccacttcg aaccggcttc caaatgcagc 120
accgagaacc ggtctaacc aaccgtggcc tcggacttgg acggcacccct cctgggtgtcc 180
cccagcgcatt ntcccttacta catgctggtc gccatcgaag ccggcagctt cttccgtggc 240
cttgccctcc ttgcctg 257

<210> 74
<211> 255
<212> DNA
<213> Glycine max

<400> 74
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gtcacggcta gtccttagggt gatggtgag ccgttggta aggctttct cggggctgac 120
aagggtctg ggactgaact tgaggccacc aaatcgggaa ctttcactgg gtttggtaag 180
aaggctggtg tgcttgggg ggagcataag aaagtggctc tggtgaagga gtttcagggt 240
aattacctga ctgg 255

<210> 75
<211> 244
<212> DNA
<213> Glycine max

<400> 75

caacaacatt cattcattca cagctgtcac gccgtgaacg gaaaatggca acggcgagac 60
gcagtttccc gccttatcacc gaatgcaacg gaacgacacc gtgcgagtc gtggccgccc 120
acctcgacgg tacgctcctc atcncccgta gctcgttccc gtacttcatg ctcgtcggc 180
tcgaagccgg cagcctcctc cgccggcctca tgcnttcctg ggtttattt gagnaccct 240
gagg 244

<210> 76
<211> 240
<212> DNA
<213> Glycine max

<400> 76
gctggctacc ctcttcttcc ccaccatcac accaatcaaa cctcactcta ccctggccat 60
ggcatggga gccttncgc cacttcgaac cggttccaa atgcagcacc gaanaccgt 120
ttnaccanac cgtggcctcg gncttggacg gcaccctcct ggtgtccct agcgcccttc 180
cttactacat gctcgtcgcc atcgaagccg gcagcttcct ccgtggcttg tcctccttgg 240

<210> 77
<211> 263
<212> DNA
<213> Glycine max

<400> 77
gtttctcggg gctgacaagg tgcttggac tgaactttag gcccaccaa at cggggacgtt 60
caactgggtt gttaagaagc ctggtgtgct tgggtggag cataagaaag tggctctgg 120
gaaggagttt cagggttaatt tacctgactt ggtcttaggt gatagtaaaa gtgattatga 180
cttcatgtca atttgcagg aagggtacat ggtgccaaga actaagtgtg aaccactacc 240
aagaaacaag ctttaagtc caa 263

<210> 78
<211> 258
<212> DNA
<213> Glycine max

<400> 78
ggccacgaaa tcggggaggt tcactgggtt tggtaaggag cctgggtgtc ttgttgggaa 60
gcacaagaaa gtggctgttgc tgaaggagtt tcagggtaat ttacctgact tgggactagg 120
agatagtaaa agtGattatg acttcatgtc aatttgcagg aagggtacat tggtgccaag 180
gactaagtgt gAACCAACTAC caagaaacaa acttttaagt ccaatttattt ntcatgaggg 240
taggtttgtt caaaggccc 258

<210> 79
<211> 260
<212> DNA
<213> Glycine max

<400> 79
ctcttcttcc ccaccatcac accaancaaa cctcacttc cctttctccc ctgaccctct 60
ccctgccatg gtcatggag ctttggca cttcgaaccg gtctccaaat gcagcaccga 120
gaaccggctt aaccaaaaccg tggcctcgga cttggacggc accctcttgg tgcctccct 180
cgcatttcct tactacatgc tggtcgccc cgaagccggc agcttcctcc gtggccctt 240
tcctccttgc cttcgccct 260

<210> 80
<211> 257
<212> DNA
<213> Glycine max

<400> 80
gggaacaaca acaaattggca ngtacccat ctccttccaa cttgggtgcatttataccctgg 60
atacccaatc cagcctgtaa ttgtacgta tcctcatgtc cactttgacc aatctgggg 120
tcatgtntct ttggaaagc ttatgttcag aatgttactt caatttcaca actttttga 180
ggtagaatat cttcctgtca tttatcccct ggtatgataag gaaactgctg tancttntcg 240
ggagaggact agccggg 257

<210> 81
<211> 272
<212> DNA
<213> Glycine max

<400> 81
catacctttt gttggcacca ttattagagc aatgcaggc atatatgtta acagattctt 60
accatcatca aggaagcagg ctgttaggaa aataaaaggaa ctgaaaaca gagaaggccc 120
tcttgtata aatttcctcg agtactatta tttcccgagg gaacaacaac taatggcagg 180
aaccttatct ccttccaact tggtgcatat atccctggat acccaatcca gcctgttaatt 240
atacgctatc ctcatgtaca ctttgaccaa tc 272

<210> 82
<211> 245
<212> DNA
<213> Glycine max

<400> 82
gggcatttca catactagag ttcatcccag tgaaaagaaa gtgggaggct gatgaatcaa 60
tcatgcgccca tatgctttct acattcaagg atccacaaga tcctctctgg cttgcgcctt 120
tcccagaagg cactgatTC actgagcaaa agtgccttcg gagtcaaaaa tatgctgctg 180
aacaataagtt accgggttctg aaaaatgttt tacttccaag gacaaagggg cttctgtgcc 240
gcttg 245

<210> 83
<211> 268
<212> DNA
<213> Glycine max

<400> 83
cagtgtcctt cctttctgga caatgtttt ggtgttgacc cttcagaagt gcacctgcac 60
gtgcggcgta ttccgggtga ggagattcca gcttctgaaa ccaaagctgc ttcttggtta 120
atcgacacat tccagatcaa ggaccaattt ctttggatt tcaagattca aggcatttc 180
cctaaccaac taaaatgaaa tggaaatttct agatTTAAGA gcctactctc ttttatggtg 240
atagtttctt ttactgccccat gtttattt 268

<210> 84
<211> 265
<212> DNA
<213> Glycine max

<400> 84
gaaagagact gggcaaaaga tggaaacatca ctgaagtcag gttttaggca tctagagcac 60
atgcattcc ctttctgggtt ggcctttttt gttgaaggaa ctcgtttcac gcagacaaag 120
cttttacaag ctcaagagtt tgctgcttca aaagggtcgc ctatacctag aaatgttttg 180
attcctcgta ctaagggttt tgcacagca gnacaaagcc ttcggccatt tcgttccagc 240
catttatgat tgcacatatg cagtt 265

<210> 85
<211> 265
<212> DNA
<213> Glycine max

<400> 85
gaaagagact gggcaaaaga tggaaacatca ctgaagtcag gttttaggca tctagagcac 60
atgcattcc ctttctgggtt ggcctttttt gttgaaggaa ctcgtttcac gcagacaaag 120
cttttacaag ctcaagagtt tgctgcttca aaagggtcgc ctatacctag aaatgttttg 180
attcctcgta ctaagggttt tgcacagca gnacaaagcc ttcggccatt tcgttccagc 240
catttatgat tgcacatatg cagtt 265

<210> 86
<211> 301
<212> DNA
<213> Zea mays

<400> 86
ctcgctgtca agggcacccc gccggccggc cccaaagaagg gccacccggg cgtcctcttc 60
gtctgcaacc accgcaccgt gctgcacccc gtcgagggtgg ccgtggcgct gcgcggcaag 120
gtcagctgcg tcacctacag catctccaag ttctccgagc tcatctcgcc catcaaggcc 180
gtcgcgtgt cgccggaggc gacaaggacg ccgagaacat ccggccctg ctggaggagg 240
gacgacgtgtt catctggccc gagggnaaca actgcccgcga gccccttctg ctgcgttcag 300
g 301

<210> 87
<211> 309

<212> DNA

<213> Zea mays

<400> 87

cgctcatgct gtgtacatca acctgccgt gcccggcgc atcgctact acaccataa 60
gctcatggcc atcaggctcg tcgtcaaggg caccggccg cccggccca agaagggcca 120
cccgccgtc ctttcgctc gcaaccaccg caccgtgctc gaccggcgtc aggtggccgt 180
ggcgtgcgc cgcaaggtca gctgcgtcac ctacagcatc tccaagttt ccgagctcat 240
ctcgccccatc aaggccgtcg cgctgtcgaa gaggcgacaa ggacgcccgg aacatccgccc 300
gcctgctgg 309

<210> 88

<211> 304

<212> DNA

<213> Zea mays

<400> 88

tggctgtgca ggaggcctac ctgggtacgt caaggaagta cagccgggtg cccaggaacc 60
agctgctgag cccgctgatt cgtcacgac ggccgcctcg tgcagcgccc gacggccgtc 120
gtcgcgtcg tcaccccttctt ctggatgccc ttccggcttcg cgctggcgtc catgcgcgtg 180
tacatcaacc tgccgctgcc cgagcgcatac gtctactaca cctacaagct catgggcatac 240
aggctcggtcg tcaagggcac cccgcccggc cccggccaaaga agggccaccc gggcgtcctc 300
ttcg 304

<210> 89

<211> 312

<212> DNA

<213> Zea mays

<400> 89

ggttcatcca cttgtgttgc tatngaccg gtaccgtagg agagcacacg actancatcg 60
caaagattt gggctacgg gacaatctcc atgttctaca atcttnaggt cgaaggaatg 120
gagaatctgc ctccaaatag ctgtcctggt gtctatgttg ctaaccatca gagcttcttg 180
gatatttata cccttctaac tcttagggagg tgcttcaaat ttataagcaa gaccagcatc 240
tttatgttcc ctattatagg gtgggcaatg tatcttcttg gtgtgattcc tctgcggcgt 300
atggacagca gg 312

<210> 90

<211> 264

<212> DNA

<213> Zea mays

<400> 90

ggtgtgttat ctgaaagaat ccacgtgtct catcaacaga aaaatgcacc aatgatgcta 60
ctctccctt gggggcacaat ctacaaatgg ggattatctc cttccattca aaacaggtgc 120
ttttcttgc aaggcaccag ttcaaccagt catttgaga tattcattaca aaagatttaa 180
tgcagcatgg gattccatgt cagggggcacc tcattgttattt ctgctgtct gtcaattttgt 240
aaattaccta gaggtggtcc gctt 264

<210> 91

<211> 212

<212> DNA

<213> Zea mays

<400> 91

aaatgtcttg gatgcatttt tggtcagcgg gagtcgaaaa caccagattt ccaaagggttt 60
tcaggtgtcg tatttggaaat aatccatcg tgcataac agaaaaatgc accaatgtg 120
ctactcttcc ctgagggcac aactacaaat ggggattatc tccttccatt caaaacaggt 180
gctttcttg ccaaaggcacc agttcaacca gt 212

<210> 92

<211> 267

<212> DNA

<213> Zea mays

<400> 92

gtctaaagaa atngaaaggc gtggggnaat tgggtctaat catgtntttt atgtggatat 60
tctttatcan atgtcagcct ctttccatg ttttgggtct aagagatcag tggntagatt 120
gcctctagtt ggtctcataa gcaaatgtc tggatgcatt tttgttcagc gggagtnnaa 180
aatncanatt tcaaagggtgt gnatctgaaa gaatccatcg tgctcatcaa 240

cagaaaaatg caccaatgat gctactc 267
<210> 93
<211> 152
<212> DNA
<213> Zea mays

<400> 93
ctacaaaatgg ggattacacctt cttccattta agactggagc cttnttgca ggtgcaccag 60
tgcagccagt cattttgaaa tacccttaca ggagatttag tccagcatgg gattcaatgg 120
atggagcacg tcatgtgtta ttgctgctct gt 152

<210> 94
<211> 274
<212> DNA
<213> Zea mays

<400> 94
aaaatataaa ttaatatggt cttaatccca ccatataaat aacgttctct ttctgcaggg 60
caatttagtt ctttctaata ttgggctggc agagaagcgc gtgtaccatg cagcaactgac 120
tggtagtagt ctacctggcg ctagacatga gaaagatgat taaaagacgt tgcgtcgctt 180
tttctgttaac agacagccga ggaacactta aaaatgttaac tgtgtgcgtg ttttatacc 240
tgtaatgtgg cagtttattt gtttgaggag gctg 274

<210> 95
<211> 295
<212> DNA
<213> Zea mays

<400> 95
aatagctatc aagtacaata aaatatggt tgatgccttt tggaaacagta agaagcaatc 60
ttttacaatg cacttggtcc ggctgtatgac atcatgggct gttgtgtgtg atgtttggta 120
cttacctcct caaatatctga gggagggaga gacggcaatt gcatttgctg agagagtaag 180
ggacatgata gctgctagag ctgactaaa gaaggttcct tggatggct atctgaaaca 240
caaccgtcct agtcccaaac acactgaaga gaacaacgca tattgccat ctgtc 295

<210> 96
<211> 273
<212> DNA
<213> Zea mays

<400> 96
gngccatctc accggcggcn ggctgcggc cgccaaccgg aggcgatggc gagctngtct 60
gtggggcgg acatggagca ntaccgcccc aacctggagg actacccccc gcccactcg 120
ctcccgcagg aggcggccag gaatctccat ctgcgcgatc tgcttgacat ctcggcgggtg 180
ctaaccgagg cagcgggtgc catagtcgat gattcattca cccgttgctt taagtcaat 240
tctccagaac catggaatgg aacatatatt tgt 273

<210> 97
<211> 127
<212> DNA
<213> Zea mays

<400> 97
ctcaaatatct ganggaggga gagactgcaa ttgcgtttgc tgagagagta agggacatga 60
tagcagctag agctggtctt aagaaggatcc cgtggatgg ctatctgaag cacaaccgccc 120
ctatcc 127

<210> 98
<211> 286
<212> DNA
<213> Zea mays

<400> 98
gaaccgtacg cgccctcatta cgcccatcca cgtgctcgcc tctcccccattc gcataatttt 60
nctccggcggc gtgcgcattc ccancggcng cnngccctgcn gccggcaacc ggaggcgatg 120
gcgagctcgat ctgtggcggc ggacatggag ctggaccgccc ccaacctggaa ggactacntc 180
ccgcccggant cgctcccgcga ggaggcgacc aggaatctcc atctgngcga tctgcttgan 240
atctcgccgg tgctaaccga ggcagcgggt gccatagtcg atgatt 286

<210> 99
<211> 308
<212> DNA
<213> Zea mays

<400> 99
cgccatctca tcggcggcgg gcgtgcggcc ggcggcngag gcgaggngcg attggcgagc 60
tcgtctgtgg cgccggacat ggagctggac cgcggccanacc tgaggacta nctcccgccc 120
gactcgnnc cgcagaggcg ccccggaatc tccanctgctg cgatctgctg gacatcnccg 180
cggtgtcac cgaggcagcg ggtgccattt tcgatgactc cttcacacgg ngcttaagt 240
caaattctcc agagccatgg aatttgaaca tatatctgtt ccccttatgt gcttttgtt 300
ataataag 308

<210> 100
<211> 282
<212> DNA
<213> Zea mays

<400> 100
cagaaactag angtagtca cagcatggca taaaattgtc atagtaaaca acancnact 60
gagcaactat gcaatttaat gccatgctgt gactaacttc tagtttctgg cattaaatta 120
ctgtttggct actaggaaga ccgaggtaga gaagcaata taagaatacc ctccaacgca 180
canccaaatg acagagtaaa tgaaggtagg gtccaccttc ttgaacatga ccgtatactg 240
gttgttaaca caagttcctc tggaaaatc agagagggtt tt 282

<210> 101
<211> 282
<212> DNA
<213> Zea mays

<400> 101
ggcgcggctg gccgtggcgc tggcctgcc gtacagtact cgacgcccgt cctggcngcg 60
acnggcgtatgt cgtggcgct caaagggtng cgcggcngc ttgcnnngcc gtgctccggc 120
ggcgctgnc agctgttcgt gtgcaacnac cggacgctga tcgacccngt gtacgtgtcc 180
gtagcgtgga ccggaaatg cgcgcgtgt nctacagnct gangcggnnt tcggagctca 240
tctccccat ngncgaaang tgcacctgan accggaaacg gg 282

<210> 102
<211> 290
<212> DNA
<213> Zea mays

<400> 102
ggacgcggca ccatgcgcgc cgagctggcc agtggcgacg tggccgtgtg ccccgagggc 60
accacgtgcc gggagccctt cctgctccgc ttctccaagc tttcgccga gctcagcgac 120
aggatcgtgc ccgtggcgat gaactaccgc gtggggctct tcacccgac gacggcgcc 180
gggtggaaag ccatggaccc catttcttc ttcatgaacn gggcccggt tacgagggtga 240
cgttcctgaa ccantccccg caaagcgacg tgcgcggcgg ggaagagccc 290

<210> 103
<211> 279
<212> DNA
<213> Zea mays

<400> 103
acgaggtgac gttcctgaac cagctccccg cagaggcgac gtgcgcggcg gggaaagagcc 60
ccgttgatgt agccaactac gttcagcgga tactcgctgc cacgctcggtt ttcgagtgc 120
ccacccctcac aagaagagac aaatacacgg tgctcgccgg caacgacggc gtcctgaacg 180
ccaagccggc ggcggcccg aagccggctt ggcagagccg cgtgaaggaa gtcctcggtt 240
tctgctccac taacaattac accttgcaca gatctggac 279

<210> 104
<211> 315
<212> DNA
<213> Zea mays

<400> 104
gcccggcgc atcgctact acacctacaa gctcatggc atcaggctcg tcgtcaaggg 60
caccggccgc cgcggccca agaagggcca cccggcgctc ctcttcgtct gcaaccaccg 120
caccgtgtc gaccccgctg aggtggccgt ggcgctgcgc cgcaangtca gtcgcgtcac 180

tacagcatct ccaagttctc cgagctcatc tcgcccattca aggccgttagc agnaaaaggcag 240
gtcgc当地 gggcggatgg gagtcgatgg aagnaaattt ggcactggtc atctgcncga 300
aggnaactg cggag 315

<210> 105

<211> 314

<212> DNA

<213> Zea mays

<400> 105

cgagacaccg agcacgtact accagcaaga tggtggcgctc tcccagattc aagccatcg 60
aggagtctg ctcggagggg cggcgagc agacgggtgc cgccgacctg gacggcacgc 120
tgctcatctc caggagcgctc ttcccctact acctccctcg ggctctcgag gccggcagcg 180
tcctccgcgc cgcgctgctc ctcctgtccg tgccgttcgt ctacgtcacc tacgccttct 240
tctccgagtc gctggccatc agcacgctgg tgtacatctc cgtggcgcccc ctcaagggtgc 300
gcanatcgag atgg 314

<210> 106

<211> 291

<212> DNA

<213> Zea mays

<400> 106

ctctgggtct gggggccgaga caccgagcac gtactaccag caagatggtg gcgtctccca 60
gattcaagcc catcgaggag tgctgctcg ggggggcggtc ggagcagacg gtggggcccg 120
acctggacgg cacgctgctc atnccagga ggcgcgttccc ctactacctc ctcgtggctc 180
tcgaggccgg cagcgtcctc cgcgcgcgc tgctgtccct gtccgtgccg ttctgtctacg 240
tcacctacgc ttctttctcc gatcgctgg ccatcagcac gctgggtgtac a 291

<210> 107

<211> 300

<212> DNA

<213> Zea mays

<400> 107

gcacgcagca gtacgacgtc tctcctctgg gtctggggcc gagacaccga gcacgtacta 60
ccagcaagat ggtgggtct cccagattca agccatcgaa ggagtgtctgc tcggaggggc 120
ggtcggagca gacgggtggcc gcccacctgg acggcacgct gctcatctcc aggagcgcgt 180
tcccctacta cctcctcggt gctctcgagg ccggcagcgt cctccgcgc ggcgtgtgc 240
tcctgtccgt gccgttcgtc tacgtcacct acgccttctt ctccgagtcg ctggccatca 300

<210> 108

<211> 284

<212> DNA

<213> Zea mays

<400> 108

gngggccgaga caccgagcac gtactaccag cangatggtg gcgtctccca gattcangcc 60
antcgaggag tgctgctcg ggggggcggtc ggagcagacg gtggccgcgc acctggacgg 120
cacgctgctc atctccagga ggcgcgttccc ctacnaccc ctcgtggctc tcgaggccgg 180
cagcgtcctc cgcgcgcgc tgctgtccct gtccgtgccg ttctgtctacg tcactacgccc 240
ttcttctccg agtgcgtggc catcaanacg ctgggtgtaca tctc 284

<210> 109

<211> 280

<212> DNA

<213> Zea mays

<400> 109

ctcctctggg tctggggccg agacaccgag cacgtactac cagcaagatg gtggcgcttc 60
ccagattcaa gcccattcgag gagtgctgtc cggaggggcg gtcggagcag acgggtggccg 120
ccgacacctggc cggcacgctg ctcatctcca ggagcgcgtt cnctactac ctcctcggtgg 180
ctctcgaggc cggcagcgtc ctccgcgcgg cgctgtgtc cctgtccgttn ccgttcgtct 240
acgtcaccta cgcntnttc tccgagtcgc tggccatcag 280

<210> 110

<211> 287

<212> DNA

<213> Zea mays

<400> 110
 cgtctctcct ctgggtctgg ggccgagaca ccgagcacgt actaccagca agatggtg 60
 gtctcccaga ttcaagccca tcgaggagtg ctgctcgag gggcggtcg agcagacgg 120
 ggccgcgcac ctggacggca gctgctcatc tccaggagcg cgttccccata ctacctcctc 180
 gtggctctcg aggccggcag cgtcctccgc gccgcgctgc tgctcctgtc cgtgccgttc 240
 gtctacgtca ctacggcttc ttctccgagt cgctggccat cagcacg 287

<210> 111
 <211> 286
 <212> DNA
 <213> Zea mays

<400> 111
 cgcacaggtta cgacgtctct cctctgggtc tggggccgag acaccgagca cgtactacca 60
 gcaagatggt ggcgtctccc agattcaagc ccatcgagga gtgctgctcg gagggcggt 120
 cggagcagac ggtggccgccc gacctggacg gcacgcgtct catctccagg agcgcgttcc 180
 cctactactc ctcgtctctc cgaggccggc aggtcctccg cgccgcgtc tgctcctgtc 240
 gtgcgttcgt cttagtacta cgcttttctc gancgtggca ataana 286

<210> 112
 <211> 323
 <212> DNA
 <213> Zea mays

<400> 112
 gttattccct gaaggtacca caacaaatgg gagattcctg atttcgttcc aacatggtgc 60
 attcatacatc ggctaccctg ttcaacctgt tttgtccgt tatccacatg tgcactttga 120
 tcaatcatgg ggnatatat cgttattaaa gctcatgttt aagatgttca cccaatttca 180
 taatttcatg gaggttagagt accttcctgt tgtctaccct cctgagatca agcaagagaa 240
 tgccttcat tttcgaggagg ataccagcta tgctatggca cgtgccctca atgtcttgcc 300
 aacttcctat tcatatggtg att 323

<210> 113
 <211> 312
 <212> DNA
 <213> Zea mays

<400> 113
 cgataaggcc ctttcgaag agcttctacc gtcggatcaa cagattctg gccgagctgc 60
 tgtggcttca gcttgtctgg gtgggtggact ggtggcagg tttaaggta caactgcatt 120
 cagatgagga aacttacaga tcaatgggta aagagcatgc actcatcata tcaaattatc 180
 ggagtgtat tgattggctc attggatggta tattggccca gcgttcagg tgccttgaa 240
 gtacacttgc tgcatacatgg aagtcatcca agttccttcc agttattggc tggtaatgt 300
 gtttgcaga gt 312

<210> 114
 <211> 279
 <212> DNA
 <213> Zea mays

<400> 114
 agtggggctt ccaaagggtt aaagacttcc ctagaccatt ttggctagct ctttttgg 60
 agggtaactcg ctttactcca gcaaagcttc tcgcagctca ggagtatgcg gcttcccagg 120
 gcttaccaggc tccttagaaat gtacttattc cacgtaccaa gggattttgtt tctgcccgtaa 180
 gtattatgcg agattttgtt ccagccattt acgatacaac tgtaatagt cctaaagatt 240
 cccctcaacc aacaatgctg cgattttga aaggccaat 279

<210> 115
 <211> 304
 <212> DNA
 <213> Zea mays

<400> 115
 cgtcaacgccc atccaggccg tcctatttgc gacgataagg ccctttcga agagcttcta 60
 ccgtcgatc aacagattct tggccgagct gctgtggctt cagttgtct ggggggtgg 120
 ctgggtggca ggtgttaagg tacaactgca tgcagatggaa gaaacttaca gatcaatggg 180
 taaagagcat gcactcatca tatcaaattca tcggagtgat attgattggc tcatggatgg 240
 atattggccc agcgttcagg gtgccttggaa agtacattgc tgcatacatgg aagtcatcca 300
 agtt 304

<210> 116
<211> 259
<212> DNA
<213> Zea mays

<400> 116
cttcctccctg tccggcctca tcgtcaacgc catccaggcc gtcctatttg tgacgataag 60
gcccnnttcg aagagctct aacgtcgat caacagatc ntggccgagc tgctgtggct 120
tcagcttgc tgggtgtgg acnngtgggc aggtgttaag gtacaactgc atgcngatga 180
gaaacttac agatcnatgg gtanagagca tgcactcatc atatcaaatc atcggagtga 240
tattgattgg cncattgga 259

<210> 117
<211> 235
<212> DNA
<213> Zea mays

<400> 117
attccacgta ccaagggatt ttttatctgct gtaagtatta tgcgagattt tgcccagcc 60
atttatgata caactgtaat agttcctaaa gattcccttc aaccaacaat gctgcggatt 120
ttgaaaggc aatcatcagt gatacatgtc cgcatgaaac gtcatgcaat gagtgagatg 180
ccaaaatcag atgaggatgt ttcaaatgg tgtaaagaca ttttgtggc aaagg 235

<210> 118
<211> 282
<212> DNA
<213> Zea mays

<400> 118
tgagatgcca aaatcagatg atgacgtttc aaaatgggt aaagacattt ttgtgacaaa 60
ggatgccta ctggacaaac atttggcaac aggacatttc gatgaggaga ttagacctat 120
cggcccccgt gtaaatcat tctgtgtac cctgtttgg tcgtgcctgc tggtgtttgg 180
tgcacatcag ttcttcaagt ggacgcagct cctatcgaca tggagaggag tggcattcac 240
tgccgcagga tggcgctcgt gacaggggtc atgcacgtct tc 282

<210> 119
<211> 166
<212> DNA
<213> Zea mays

<400> 119
ctgggtggca ggcgttaagg tacaactaca tgcggatgag gacacttacc gatcaatggg 60
taaagagcat gcactcgtca tatcaaata tcgaagtgtat attgattggc ttattggatg 120
gatattggcc cagcgctcag ggtgccttgg aagtacgctc gctgtc 166

<210> 120
<211> 234
<212> DNA
<213> Zea mays

<400> 120
agtcanccaa gntccttcca gtcattggct ggtcaatgtg gtttgcagag tacccctttt 60
nggagaggag ctgggccaag gatgaaaaga cactaaatgt gggctccaa aggttgaag 120
actcccttag accattnng ctagctctt tttgtngagg gnantcgct tactccagca 180
angntntng aggnncagn agnnncgggn ttcccanggg ttaacagncc cana 234

<210> 121
<211> 210
<212> DNA
<213> Zea mays

<400> 121
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atccggccgccc cagtgaatc atngctgggt accctgtntt ggtcgtgcct gctgttgc 180
ggtgccatcg agntcttcaa gtggacgcag 210

<210> 122
<211> 274
<212> DNA

<213> Zea mays

<400> 122

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tctgaccct ccgagatcgn aagcggcgcc catggcgatc cgcgtcgatc tcgtcggtct 180
cccgtcggc ctcctctcc tcctgtccgg cctcatcgatc aacaccatcc aggccatcct 240
attttgaca ataaggccct ttccaagag ctta 274

<210> 123

<211> 305

<212> DNA

<213> Zea mays

<400> 123

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gggagattga tgaagcaatt attcagaaca agctatcaa atttaagaac ccgagagatc 180
ctatctggtt ggccgtttt cctgaaggca cggattatac tgagaagaaa tgcatcatga 240
gtcaagatc tgcttcagaa catggcttgc ctatgtatc acatgtccctc cttccaaaga 300
caagg 305

<210> 124

<211> 279

<212> DNA

<213> Zea mays

<400> 124

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aggttaggc agaaggacca gtcctggca gatttcttca tgaagggca tttctgtatg 180
aaaggaactg aaaggagatc tgcacgccc gagtgcctgg caaactttttaaaccaggtag 240
tatgcttgc ggccnatctg gtttgtacct aaactctt 279

<210> 125

<211> 219

<212> DNA

<213> Zea mays

<400> 125

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gtttaggcag aaggaccagc tctggcaga tttcttcatg aaggggact ttcctgtatg 180
aggaactgaa ggagatctgt cgacgcccgaat gtgcctggc 219

<210> 126

<211> 293

<212> DNA

<213> Zea mays

<400> 126

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ctccgacata ncggcgtccg aaaaacgggg tggctggcng gtnnnngtggaa gcggttcaag 180
gcntnganna acgactngc ttttccatcgc ggtctggggcc aatttcnccc 240
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<211> 6

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<210> 128

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<400> 128

Val Thr Tyr Ser Xaa Ser
1 5

<210> 129

<211> 7

<212> PRT

<213> conserved sequence

<400> 129

Val Xaa Leu Thr Arg Xaa Arg
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<210> 130

<211> 5

<212> PRT

<213> conserved sequence

<400> 130

Cys Pro Glu Gly Thr
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<210> 131

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<213> conserved sequence

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Ile Val Pro Val Ala
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<210> 132

<211> 7

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<400> 132

Leu Xaa Xaa Gly Asp Leu Val
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<210> 133

<211> 6

<212> PRT

<213> conserved sequence

<400> 133

Phe Xaa Xaa Gly Ala Phe
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<210> 134

<211> 6

<212> PRT

<213> Synthetic Oligonucleotide

<400> 134

Val Ala Asn Xaa Xaa Gln
1 5

<210> 135

<211> 30

<212> DNA

<213> Synthetic Oligonucleotide
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<400> 137
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<210> 147
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<210> 152
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<210> 153
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<210> 154
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<210> 156
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<210> 157
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<400> 157
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<210> 158
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<400> 158
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<210> 159
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<210> 160
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<213> Synthetic Oligonucleotide

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<213> Simmondsia chinensis

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1702

<210> 162

<211> 387

<212> PRT

<213> *Simmondsia chinensis*

<400> 162

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Leu Val Asp

Val Clu I

Val Giu L
E0

See Vol. I

Ser Val L

— 2 —

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115 120 125

Trp Ser Met Trp Phe Ser Glu Tyr Leu Phe Leu Glu Arg Ser Trp Ala
130 135 140

Lys Asp Glu Ser Thr Leu Lys Leu Gly Leu Gln Arg Leu Lys Asp Tyr
145 150 155 160

Pro Leu Pro Phe Trp Leu Ala Leu Phe Val Glu Gly Thr Arg Phe Thr
165 170 175

Gln Ala Lys Leu Leu Ala Ala Gln Glu Tyr Ala Thr Ser Met Gly Leu
180 185 190

Pro Val Pro Arg Asn Thr Leu Ile Pro Arg Thr Lys Gly Phe Val Ser
195 200 205

Ala Val Ser His Met Arg Ser Phe Val Pro Ala Ile Tyr Asp Val Thr
210 215 220

Val Ala Ile Pro Lys Ser Ser Ser Gln Pro Thr Met Leu Arg Leu Phe
225 230 235 240

Lys Gly Gln Pro Ser Thr Val His Val His Ile Lys Arg Arg Ser Met
245 250 255

Lys Asp Leu Pro Glu Ala Ala Asp Asp Val Ala Gln Trp Cys Arg Asp
260 265 270

Thr Phe Val Ala Lys Asp Ala Leu Leu Asp Lys His Asn Val Asp Asp
275 280 285

Thr Phe Gly Asp Glu Tyr Leu Gln Asp Thr Gly Arg Pro Leu Lys Ser
290 295 300

Leu Phe Val Ala Val Ser Trp Ala Leu Ile Leu Ile Leu Gly Gly Leu
305 310 315 320

Lys Phe Leu Arg Trp Ser Ser Leu Leu Ser Ser Trp Lys Gly Val Ala
325 330 335

Phe Ser Ala Ala Cys Leu Val Leu Val Thr Ile Leu Met Gln Ile Leu
340 345 350

Ile Gln Phe Ser Gln Ser Glu Arg Ser Thr Pro Ala Lys Val Ala Pro
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Gly Lys Pro Lys Asn Met Val Ser Glu Pro Thr Glu Thr Gln Arg His
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Lys Gln His
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<211> 43

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<220>

<223> Description of Artificial Sequence: Synthetic
Oligonucleotide

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<210> 164

<211> 35

<212> DNA
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<210> 165
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<213> Artificial Sequence

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<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 165
ggatccgcgg ccgcacaatg acgagctta ctactccct tcat 44

<210> 166
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<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 166
ggatccccctg caggtagag atccattgtat tctgcaat 38

<210> 167
<211> 38
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 167
ggatccgcgg ccgcataatg gaatcagagc tcaaagat 38

<210> 168
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<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 168
ggatccccctg caggcattc ttctttctga tggaaatc 38

<210> 169
<211> 41
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

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ggatccgcgg ccgcacaatg actcgttcac aagatgttca 41

<210> 170
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<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 170
ggatccccctg caggtcactt ctcttccaaat cttagccag 38

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<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 171
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<220>
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<400> 172
ggatccccctg caggttattt tttcttgaca actccgttat taccgg 46

<210> 173
<211> 39
<212> DNA
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<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 173
atatccgcgg ccgcacaatg gttatggagc aagctggaa 39

<210> 174
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<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 174
ggatccccctg caggtcaatg gagacaaggc tcgaaaagt 38

<210> 175
<211> 42
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 175

ggatccgcgg ccgcacaatg tccgccaaga tttcaatatt cc 42
<210> 176
<211> 38
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 176
ggatccccctg caggttaatt tttcttaact actccatt 38
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<210> 178
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<400> 178
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<210> 179
<211> 44
<212> DNA
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<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 179
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<210> 180
<211> 44
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 180
ggatccccctg caggttatgt tggggccaag tcaggtgcaa agat 44
<210> 181
<211> 44
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 181
ggatccgcgg ccgcaaaatg gaaaaaaaaaga gtgtacccaa ttct 44
<210> 182
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<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 182
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<210> 183
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<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 183
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<400> 184
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<210> 185
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<220>
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<400> 185
ggatccgcgg ccgcacaatg tcttttaggg atgtcctag 39
<210> 186
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<223> Description of Artificial Sequence:Synthetic Oligonucleotide

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<210> 188
<211> 60
<212> DNA
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<400> 188
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<210> 189
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<400> 189
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<210> 190
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<210> 191
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<210> 192
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<223> Description of Artificial Sequence:Synthetic Oligonucleotide

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<400> 193
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<210> 194
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<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 194
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<211> 60
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 195
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<210> 196
<211> 60
<212> DNA
<213> Artificial Sequence

<220>
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<400> 196
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<210> 197
<211> 44
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 197
ggatccgcgg ccgcacaatg gaaaagtaca ccaattggag agac 44

<210> 198
<211> 42
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 198
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<210> 199
<211> 60

<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 199
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<211> 60
<212> DNA
<213> Artificial Sequence

<220>
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<400> 200
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<210> 201
<211> 41
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 201
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<210> 202
<211> 38
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 202
ggatccccctg caggctacgc atctccttct ttccccttc 38

<210> 203
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<210> 204
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<212> DNA
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<220>
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<400> 204
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<400> 205
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<210> 207
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<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 207
atgtctgctc ccgctgccga tcataacgct gccaaaccta ctgtgcggta tttcacaccg 60

<210> 208
<211> 60
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 208
tcattcttcc tttcgtgtt ctctttctg tcttaccaggc agattgtact gagagtgcac 60

<210> 209
<211> 49
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 209
ggatccgcgg ccgcacaatg ctgcataaaa aaatagctca taaagttcg 49

<210> 210
<211> 49
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 210

ggatccccctg caggtcaaaa aataaaaacaa taaagtttat aaactaacc 49

<210> 211
<211> 60
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 211
atgctgcatt aaaaaatagc tcataaaagtt cgaaaagtccg ctgtgcggta tttcacacccg 60

<210> 212
<211> 60
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 212
tcaaaaaata aaacaataaa gtttataaac taaccaaatt agattgtact gagagtgcac 60

<210> 213
<211> 41
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 213
ggatccgcgg ccgcacaatg agtgtgatag gtaggttctt g 41

<210> 214
<211> 41
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 214
ggatccccctg caggtaatg catttttt acagatgaac c 41

<210> 215
<211> 60
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 215
atgagtgtga tagtaggtt cttgttattac ttgaggtccg ctgtgcggta tttcacacccg 60

<210> 216
<211> 60
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Synthetic Oligonucleotide

<400> 216
ttaatgcac tttttacag atgaaccttc gttatggta agattgtact gagagtgcac 60

<210> 217

<211> 381

<212> PRT

<213> *Saccharomyces* sp.

<220>

<400> 217

Met Ser Phe Arg Asp Val Leu Glu Arg Gly Asp Glu Phe Leu Glu Ala
1 5 10 15

Tyr Pro Arg Arg Ser Pro Leu Trp Arg Phe Leu Ser Tyr Ser Thr Ser
20 25 30

Leu Leu Thr Phe Gly Val Ser Lys Leu Leu Leu Phe Thr Cys Tyr Asn
35 40 45

Val Lys Leu Asn Gly Phe Glu Lys Leu Glu Thr Ala Leu Glu Arg Ser
50 55 60

Lys Arg Glu Asn Arg Gly Leu Met Thr Val Met Asn His Met Ser Met
65 70 75 80

Val Asp Asp Pro Leu Val Trp Ala Thr Leu Pro Tyr Lys Leu Phe Thr
85 90 95

Ser Leu Asp Asn Ile Arg Trp Ser Leu Gly Ala His Asn Ile Cys Phe
100 105 110

Gln Asn Lys Phe Leu Ala Asn Phe Phe Ser Leu Gly Gln Val Leu Ser
115 120 125

Thr Glu Arg Phe Gly Val Gly Pro Phe Gln Gly Ser Ile Asp Ala Ser
130 135 140

Ile Arg Leu Leu Ser Pro Asp Asp Thr Leu Asp Leu Glu Trp Thr Pro
145 150 155 160

His Ser Glu Val Ser Ser Ser Leu Lys Lys Ala Tyr Ser Pro Pro Ile
165 170 175

Ile Arg Ser Lys Pro Ser Trp Val His Val Tyr Pro Glu Gly Phe Val
180 185 190

Leu Gln Leu Tyr Pro Pro Phe Glu Asn Ser Met Arg Tyr Phe Lys Trp
195 200 205

Gly Ile Thr Arg Met Ile Leu Glu Ala Thr Lys Pro Pro Ile Val Val
210 215 220

Pro Ile Phe Ala Thr Gly Phe Glu Lys Ile Ala Ser Glu Ala Val Thr
225 230 235 240

Asp Ser Met Phe Arg Gln Ile Leu Pro Arg Asn Phe Gly Ser Glu Ile
245 250 255

Asn Val Thr Ile Gly Asp Pro Leu Asn Asp Asp Leu Ile Asp Arg Tyr
260 265 270

Arg Lys Glu Trp Thr His Leu Val Glu Lys Tyr Tyr Asp Pro Lys Asn
275 280 285

Pro Asn Asp Leu Ser Asp Glu Leu Lys Tyr Gly Lys Glu Ala Gln Asp
290 295 300

Leu Arg Ser Arg Leu Ala Ala Glu Leu Arg Ala His Val Ala Glu Ile

305	310	315	320
Arg Asn Glu Val Arg Lys Leu Pro Arg Glu Asp Pro Arg Phe Lys Ser			
325 330 335			
Pro Ser Trp Trp Lys Arg Phe Asn Thr Thr Glu Gly Lys Ser Asp Pro			
340 345 350			
Asp Val Lys Val Ile Gly Glu Asn Trp Ala Ile Arg Arg Met Gln Lys			
355 360 365			
Phe Leu Pro Pro Glu Gly Lys Pro Lys Gly Lys Asp Asp			
370 375 380			

<210> 218

<211> 396

<212> PRT

<213> *Saccharomyces* sp.

<220>

<400>. 218

Met Lys His Ser Gln Lys Tyr Arg Arg Tyr Gly Ile Tyr Glu Lys Thr			
1	5	10	15

Gly Asn Pro Phe Ile Lys Gly Leu Gln Arg Leu Leu Ile Ala Cys Leu		
20	25	30

Phe Ile Ser Gly Ser Leu Ser Ile Val Val Phe Gln Ile Cys Leu Gln		
35	40	45

Val Leu Leu Pro Trp Ser Lys Ile Arg Phe Gln Asn Gly Ile Asn Gln		
50	55	60

Ser Lys Lys Ala Phe Ile Val Leu Leu Cys Met Ile Leu Asn Met Val			
65	70	75	80

Ala Pro Ser Ser Leu Asn Val Thr Phe Glu Thr Ser Arg Pro Leu Lys		
85	90	95

Asn Ser Ser Asn Ala Lys Pro Cys Phe Arg Phe Lys Asp Arg Ala Ile		
100	105	110

Ile Ile Ala Asn His Gln Met Tyr Ala Asp Trp Ile Tyr Leu Trp Trp		
115	120	125

Leu Ser Phe Val Ser Asn Leu Gly Gly Asn Val Tyr Ile Ile Leu Lys		
130	135	140

Lys Ala Leu Gln Tyr Ile Pro Leu Leu Gly Phe Gly Met Arg Asn Phe			
145	150	155	160

Lys Phe Ile Phe Leu Ser Arg Asn Trp Gln Lys Asp Glu Lys Ala Leu		
165	170	175

Thr Asn Ser Leu Val Ser Met Asp Leu Asn Ala Arg Cys Lys Gly Pro		
180	185	190

Leu Thr Asn Tyr Lys Ser Cys Tyr Ser Lys Thr Asn Glu Ser Ile Ala		
195	200	205

Ala Tyr Asn Leu Ile Met Phe Pro Glu Gly Thr Asn Leu Ser Leu Lys		
210	215	220

Thr Arg Glu Lys Ser Glu Ala Phe Cys Gln Arg Ala His Leu Asp His			
225	230	235	240

Val Gln Leu Arg His Leu Leu Leu Pro His Ser Lys Gly Leu Lys Phe		
245	250	255

Ala Val Glu Lys Leu Ala Pro Ser Leu Asp Ala Ile Tyr Asp Val Thr
 260 265 270
 Ile Gly Tyr Ser Pro Ala Leu Arg Thr Glu Tyr Val Gly Thr Lys Phe
 275 280 285
 Thr Leu Lys Lys Ile Phe Leu Met Gly Val Tyr Pro Glu Lys Val Asp
 290 295 300
 Phe Tyr Ile Arg Glu Phe Arg Val Asn Glu Ile Pro Leu Gln Asp Asp
 305 310 315 320
 Glu Val Phe Phe Asn Trp Leu Leu Gly Val Trp Lys Glu Lys Asp Gln
 325 330 335
 Leu Leu Glu Asp Tyr Tyr Asn Thr Gly Gln Phe Lys Ser Asn Ala Lys
 340 345 350
 Asn Asp Asn Gln Ser Ile Val Val Thr Thr Gln Thr Thr Gly Phe Gln
 355 360 365
 His Glu Thr Leu Thr Pro Arg Ile Leu Ser Tyr Tyr Gly Phe Phe Ala
 370 375 380
 Phe Leu Ile Leu Val Phe Val Met Lys Lys Asn His
 385 390 395

<210> 219

<211> 479

<212> PRT

<213> *Saccharomyces* sp.

<220>

<400> 219
 Met Gly Phe Val Asp Phe Phe Glu Thr Tyr Met Val Gly Ser Arg Val
 1 5 10 15
 Gln Phe Lys Gln Leu Asp Ile Ser Asp Trp Leu Ser Leu Thr Pro Arg
 20 25 30
 Leu Leu Ile Leu Phe Gly Tyr Phe Tyr Leu His Ser Phe Phe Thr Ala
 35 40 45
 Ile Asn Gln Phe Leu Gln Phe Ile Asn Thr Asn Ser Phe Cys Leu Arg
 50 55 60
 Leu His Leu Leu Tyr Asp Arg Phe Trp Ser His Val Pro Ile Ile Gly
 65 70 75 80
 Glu Tyr Lys Ile Arg Leu Leu Ser Arg Ala Leu Thr Tyr Ser Lys Leu
 85 90 95
 Lys Ile Ile Pro Thr Leu Asp Lys Val Leu Glu Ala Ile Glu Ile Trp
 100 105 110
 Phe Gln Leu His Leu Val Glu Met Thr Phe Glu Lys Lys Lys Asn Val
 115 120 125
 Gln Ile Phe Ile Thr Glu Gly Ser Asp Asp Leu Asn Phe Phe Lys Asp
 130 135 140
 Ser Lys Phe Gln Thr Thr Leu Met Ile Cys Asn His Arg Ser Val Asn
 145 150 155 160
 Asp Tyr Thr Leu Ile Asn Tyr Leu Phe Leu Lys Ser Cys Pro Thr Lys
 165 170 175

Phe Tyr Thr Lys Trp Glu Phe Leu Gln Lys Leu Arg Lys Gly Glu Asp
 180 185 190

Leu Ala Glu Trp Pro Gln Leu Lys Phe Leu Gly Trp Gly Lys Met Phe
 195 200 205

Asn Phe Pro Arg Leu Asp Leu Leu Lys Asn Ile Phe Phe Lys Asp Glu
 210 215 220

Thr Leu Ala Leu Ser Ser Asn Glu Leu Arg Asp Ile Leu Glu Arg Gln
 225 230 235 240

Asn Asn Gln Ala Ile Thr Ile Phe Pro Glu Val Asn Ile Met Ser Leu
 245 250 255

Glu Leu Ser Ile Ile Gln Arg Lys Leu His Gln Asp Phe Pro Phe Val
 260 265 270

Ile Asn Phe Tyr Asn Leu Leu Tyr Pro Arg Phe Lys Asn Phe Thr Thr
 275 280 285

Leu Met Ala Ala Phe Ser Ser Ile Lys Asn Ile Lys Arg Lys Lys Asn
 290 295 300

Arg Asn Asn Ile Ile Lys Glu Ala Arg Tyr Leu Phe His Arg Glu Leu
 305 310 315 320

Asp Lys Leu Val His Lys Ser Met Lys Met Glu Ser Ser Lys Val Ser
 325 330 335

Asp Lys Thr Thr Pro Pro Met Ile Val Asp Asn Ser Tyr Leu Leu Thr
 340 345 350

Lys Lys Glu Glu Ile Ser Ser Gly Lys Pro Lys Val Val Arg Ile Asn
 355 360 365

Pro Tyr Ile Tyr Asp Val Thr Ile Ile Tyr Tyr Arg Val Lys Tyr Thr
 370 375 380

Asp Ser Gly His Asp His Thr Asn Gly Asp Leu Arg Leu His Lys Gly
 385 390 395 400

Tyr Gln Leu Glu Gln Ile Ser Pro Thr Ile Phe Glu Met Ile Gln Pro
 405 410 415

Glu Met Glu Ser Glu Asn Asn Ile Lys Asp Lys Asp Pro Ile Val Val
 420 425 430

Met Val Asn Val Lys Lys His Gln Ile Gln Pro Leu Leu Ala Tyr Asn
 435 440 445

Asp Glu Ser Leu Glu Lys Trp Leu Glu Asn Arg Trp Ile Glu Lys Asp
 450 455 460

Arg Leu Ile Glu Ser Leu Gln Lys Asn Ile Lys Ile Glu Thr Lys
 465 470 475

<210> 220

<211> 300

<212> PRT

<213> *Saccharomyces* sp.

<400> 220

Met Glu Lys Tyr Thr Asn Trp Arg Asp Asn Gly Thr Gly Ile Ala Pro
 1 5 10 15Phe Leu Pro Asn Thr Ile Arg Lys Pro Ser Lys Val Met Thr Ala Cys
 20 25 30

Leu Leu Gly Ile Leu Gly Val Lys Thr Ile Ile Met Leu Pro Leu Ile
 35 40 45
 Met Leu Tyr Leu Leu Thr Gly Gln Asn Asn Leu Leu Gly Leu Ile Leu
 50 55 60
 Lys Phe Thr Phe Ser Trp Lys Glu Glu Ile Thr Val Gln Gly Ile Lys
 65 70 75 80
 Lys Arg Asp Val Arg Lys Ser Lys His Tyr Pro Gln Lys Gly Lys Leu
 85 90 95
 Tyr Ile Cys Asn Cys Thr Ser Pro Leu Asp Ala Phe Ser Val Val Leu
 100 105 110
 Leu Ala Gln Gly Pro Val Thr Leu Leu Val Pro Ser Asn Asp Ile Val
 115 120 125
 Tyr Lys Val Ser Ile Arg Glu Phe Ile Asn Phe Ile Leu Ala Gly Gly
 130 135 140
 Leu Asp Ile Lys Leu Tyr Gly His Glu Val Ala Glu Leu Ser Gln Leu
 145 150 155 160
 Gly Asn Thr Val Asn Phe Met Phe Ala Glu Gly Thr Ser Cys Asn Gly
 165 170 175
 Lys Ser Val Leu Pro Phe Ser Ile Thr Gly Lys Lys Leu Lys Glu Phe
 180 185 190
 Ile Asp Pro Ser Ile Thr Thr Met Asn Pro Ala Met Ala Lys Thr Lys
 195 200 205
 Lys Phe Glu Leu Gln Thr Ile Gln Ile Lys Thr Asn Lys Thr Ala Ile
 210 215 220
 Thr Thr Leu Pro Ile Ser Asn Met Glu Tyr Leu Ser Arg Phe Leu Asn
 225 230 235 240
 Lys Gly Ile Asn Val Lys Cys Lys Ile Asn Glu Pro Gln Val Leu Ser
 245 250 255
 Asp Asn Leu Glu Glu Leu Arg Val Ala Leu Asn Gly Gly Asp Lys Tyr
 260 265 270
 Lys Leu Val Ser Arg Lys Leu Asp Val Glu Ser Lys Arg Asn Phe Val
 275 280 285
 Lys Glu Tyr Ile Ser Asp Gln Arg Lys Lys Arg Lys
 290 295 300

<210> 221
 <211> 759
 <212> PRT
 <213> *Saccharomyces* sp.

<400> 221
 Met Pro Ala Pro Lys Leu Thr Glu Lys Phe Ala Ser Ser Lys Ser Thr
 1 5 10 15
 Gln Lys Thr Thr Asn Tyr Ser Ser Ile Glu Ala Lys Ser Val Lys Thr
 20 25 30
 Ser Ala Asp Gln Ala Tyr Ile Tyr Gln Glu Pro Ser Ala Thr Lys Lys
 35 40 45
 Ile Leu Tyr Ser Ile Ala Thr Trp Leu Leu Tyr Asn Ile Phe His Cys
 50 55 60

Phe Phe Arg Glu Ile Arg Gly Arg Gly Ser Phe Lys Val Pro Gln Gln
 65 70 75 80

Gly Pro Val Ile Phe Val Ala Ala Pro His Ala Asn Gln Phe Val Asp
 85 90 95

Pro Val Ile Leu Met Gly Glu Val Lys Lys Ser Val Asn Arg Arg Val
 100 105 110

Ser Phe Leu Ile Ala Glu Ser Ser Leu Lys Gln Pro Pro Ile Gly Phe
 115 120 125

Leu Ala Ser Phe Phe Met Ala Ile Gly Val Val Arg Pro Gln Asp Asn
 130 135 140

Leu Lys Pro Ala Glu Gly Thr Ile Arg Val Asp Pro Thr Asp Tyr Lys
 145 150 155 160

Arg Val Ile Gly His Asp Thr His Phe Leu Thr Asp Cys Met Pro Lys
 165 170 175

Gly Leu Ile Gly Leu Pro Lys Ser Met Gly Phe Gly Glu Ile Gln Ser
 180 185 190

Ile Glu Ser Asp Thr Ser Leu Thr Leu Arg Lys Glu Phe Lys Met Ala
 195 200 205

Lys Pro Glu Ile Lys Thr Ala Leu Leu Thr Gly Thr Thr Tyr Lys Tyr
 210 215 220

Ala Ala Lys Val Asp Gln Ser Cys Val Tyr His Arg Val Phe Glu His
 225 230 235 240

Leu Ala His Asn Asn Cys Ile Gly Ile Phe Pro Glu Gly Ser His
 245 250 255

Asp Arg Thr Asn Leu Leu Pro Leu Lys Ala Gly Val Ala Ile Met Ala
 260 265 270

Leu Gly Cys Met Asp Lys His Pro Asp Val Asn Val Lys Ile Val Pro
 275 280 285

Cys Gly Met Asn Tyr Phe His Pro His Lys Phe Arg Ser Arg Ala Val
 290 295 300

Val Glu Phe Gly Asp Pro Ile Glu Ile Pro Lys Glu Leu Val Ala Lys
 305 310 315 320

Tyr His Asn Pro Glu Thr Asn Arg Asp Ala Val Lys Glu Leu Leu Asp
 325 330 335

Thr Ile Ser Lys Gly Leu Gln Ser Val Thr Val Thr Cys Ser Asp Tyr
 340 345 350

Glu Thr Leu Met Val Val Gln Thr Ile Arg Arg Leu Tyr Met Thr Gln
 355 360 365

Phe Ser Thr Lys Leu Pro Leu Pro Leu Ile Val Glu Met Asn Arg Arg
 370 375 380

Met Val Lys Gly Tyr Glu Phe Tyr Arg Asn Asp Pro Lys Ile Ala Asp
 385 390 395 400

Leu Thr Lys Asp Ile Met Ala Tyr Asn Ala Ala Leu Arg His Tyr Asn
 405 410 415

Leu Pro Asp His Leu Val Glu Glu Ala Lys Val Asn Phe Ala Lys Asn
 420 425 430

Leu Gly Leu Val Phe Phe Arg Ser Ile Gly Leu Cys Ile Leu Phe Ser
 435 440 445
 Leu Ala Met Pro Gly Ile Ile Met Phe Ser Pro Val Phe Ile Leu Ala
 450 455 460
 Lys Arg Ile Ser Gln Glu Lys Ala Arg Thr Ala Leu Ser Lys Ser Thr
 465 470 475 480
 Val Lys Ile Lys Ala Asn Asp Val Ile Ala Thr Trp Lys Ile Leu Ile
 485 490 495
 Gly Met Gly Phe Ala Pro Leu Leu Tyr Ile Phe Trp Ser Val Leu Ile
 500 505 510
 Thr Tyr Tyr Leu Arg His Lys Pro Trp Asn Lys Ile Tyr Val Phe Ser
 515 520 525
 Gly Ser Tyr Ile Ser Cys Val Ile Val Thr Tyr Ser Ala Leu Ile Val
 530 535 540
 Gly Asp Ile Gly Met Asp Gly Phe Lys Ser Leu Arg Pro Leu Val Leu
 545 550 555 560
 Ser Leu Thr Ser Pro Lys Gly Leu Gln Lys Leu Gln Lys Asp Arg Arg
 565 570 575
 Asn Leu Ala Glu Arg Ile Ile Glu Val Val Asn Asn Phe Gly Ser Glu
 580 585 590
 Leu Phe Pro Asp Phe Asp Ser Ala Ala Leu Arg Glu Glu Phe Asp Val
 595 600 605
 Ile Asp Glu Glu Glu Glu Asp Arg Lys Thr Ser Glu Leu Asn Arg Arg
 610 615 620
 Lys Met Leu Arg Lys Gln Lys Ile Lys Arg Gln Glu Lys Asp Ser Ser
 625 630 635 640
 Ser Pro Ile Ile Ser Gln Arg Asp Asn His Asp Ala Tyr Glu His His
 645 650 655
 Asn Gln Asp Ser Asp Gly Val Ser Leu Val Asn Ser Asp Asn Ser Leu
 660 665 670
 Ser Asn Ile Pro Leu Phe Ser Ser Thr Phe His Arg Lys Ser Glu Ser
 675 680 685
 Ser Leu Ala Ser Thr Ser Val Ala Pro Ser Ser Ser Glu Phe Glu
 690 695 700
 Val Glu Asn Glu Ile Leu Glu Glu Lys Asn Gly Leu Ala Ser Lys Ile
 705 710 715 720
 Ala Gln Ala Val Leu Asn Lys Arg Ile Gly Glu Asn Thr Ala Arg Glu
 725 730 735
 Glu
 740 745 750
 Glu Gly Lys Glu Gly Asp Ala
 755

<210> 222

<211> 743

<212> PRT

<213> Saccharomyces sp.

<400> 222

Met Ser Ala Pro Ala Ala Asp His Asn Ala Ala Lys Pro Ile Pro His
1 5 10 15

Val Pro Gln Ala Ser Arg Arg Tyr Lys Asn Ser Tyr Asn Gly Phe Val
20 25 30

Tyr Asn Ile His Thr Trp Leu Tyr Asp Val Ser Val Phe Leu Phe Asn
35 40 45

Ile Leu Phe Thr Ile Phe Phe Arg Glu Ile Lys Val Arg Gly Ala Tyr
50 55 60

Asn Val Pro Glu Val Gly Val Pro Thr Ile Leu Val Cys Ala Pro His
65 70 75 80

Ala Asn Gln Phe Ile Asp Pro Ala Leu Val Met Ser Gln Thr Arg Leu
85 90 95

Leu Lys Thr Ser Ala Gly Lys Ser Arg Ser Arg Met Pro Cys Phe Val
100 105 110

Thr Ala Glu Ser Ser Phe Lys Lys Arg Phe Ile Ser Phe Phe Gly His
115 120 125

Ala Met Gly Gly Ile Pro Val Pro Arg Ile Gln Asp Asn Leu Lys Pro
130 135 140

Val Asp Glu Asn Leu Glu Ile Tyr Ala Pro Asp Leu Lys Asn His Pro
145 150 155 160

Glu Ile Ile Lys Gly Arg Ser Lys Asn Pro Gln Thr Thr Pro Val Asn
165 170 175

Phe Thr Lys Arg Phe Ser Ala Lys Ser Leu Leu Gly Leu Pro Asp Tyr
180 185 190

Leu Ser Asn Ala Gln Ile Lys Glu Ile Pro Asp Asp Glu Thr Ile Ile
195 200 205

Leu Ser Ser Pro Phe Arg Thr Ser Lys Ser Lys Val Val Glu Leu Leu
210 215 220

Thr Asn Gly Thr Asn Phe Lys Tyr Ala Glu Lys Ile Asp Asn Thr Glu
225 230 235 240

Thr Phe Gln Ser Val Phe Asp His Leu His Thr Lys Gly Cys Val Gly
245 250 255

Ile Phe Pro Glu Gly Gly Ser His Asp Arg Pro Ser Leu Leu Pro Ile
260 265 270

Lys Ala Gly Val Ala Ile Met Ala Leu Gly Ala Val Ala Ala Asp Pro
275 280 285

Thr Met Lys Val Ala Val Val Pro Cys Gly Leu His Tyr Phe His Arg
290 295 300

Asn Lys Phe Arg Ser Arg Ala Val Leu Glu Tyr Gly Glu Pro Ile Val
305 310 315 320

Val Asp Gly Lys Tyr Gly Glu Met Tyr Lys Asp Ser Pro Arg Glu Thr
325 330 335

Val Ser Lys Leu Leu Lys Ile Thr Asn Ser Leu Phe Ser Val Thr
340 345 350

Glu Asn Ala Pro Asp Tyr Asp Thr Leu Met Val Ile Gln Ala Ala Arg
355 360 365

Arg Leu Tyr Gln Pro Val Lys Val Arg Leu Pro Leu Pro Ala Ile Val

370	375	380
Glu Ile Asn Arg Arg Leu Leu Phe Gly Tyr Ser Lys Phe Lys Asp Asp		
385	390	395 400
Pro Arg Ile Ile His Leu Lys Lys Leu Val Tyr Asp Tyr Asn Arg Lys		
405	410	415
Leu Asp Ser Val Gly Leu Lys Asp His Gln Val Met Gln Leu Lys Thr		
420	425	430
Thr Lys Leu Glu Ala Leu Arg Cys Phe Val Thr Leu Ile Val Arg Leu		
435	440	445
Ile Lys Phe Ser Val Phe Ala Ile Leu Ser Leu Pro Gly Ser Ile Leu		
450	455	460
Phe Thr Pro Ile Phe Ile Ile Cys Arg Val Tyr Ser Glu Lys Lys Ala		
465	470	475 480
Lys Glu Gly Leu Lys Lys Ser Leu Val Lys Ile Lys Gly Thr Asp Leu		
485	490	495
Leu Ala Thr Trp Lys Leu Ile Val Ala Leu Ile Leu Ala Pro Ile Leu		
500	505	510
Tyr Val Thr Tyr Ser Ile Leu Leu Ile Ile Leu Ala Arg Lys Gln His		
515	520	525
Tyr Cys Arg Ile Trp Val Pro Ser Asn Asn Ala Phe Ile Gln Phe Val		
530	535	540
Tyr Phe Tyr Ala Leu Leu Val Phe Thr Thr Tyr Ser Ser Leu Lys Thr		
545	550	555 560
Gly Glu Ile Gly Val Asp Leu Phe Lys Ser Leu Arg Pro Leu Phe Val		
565	570	575
Ser Ile Val Tyr Pro Gly Lys Lys Ile Glu Glu Ile Gln Thr Thr Arg		
580	585	590
Lys Asn Leu Ser Leu Glu Leu Thr Ala Val Cys Asn Asp Leu Gly Pro		
595	600	605
Leu Val Phe Pro Asp Tyr Asp Lys Leu Ala Thr Glu Ile Phe Ser Lys		
610	615	620
Arg Asp Gly Tyr Asp Val Ser Ser Asp Ala Glu Ser Ser Ile Ser Arg		
625	630	635 640
Met Ser Val Gln Ser Arg Ser Arg Ser Ser Ile His Ser Ile Gly		
645	650	655
Ser Leu Ala Ser Asn Ala Leu Ser Arg Val Asn Ser Arg Gly Ser Leu		
660	665	670
Thr Asp Ile Pro Ile Phe Ser Asp Ala Lys Gln Gly Gln Trp Lys Ser		
675	680	685
Glu Gly Glu Thr Ser Glu Asp Glu Asp Glu Phe Asp Glu Lys Asn Pro		
690	695	700
Ala Ile Val Gln Thr Ala Arg Ser Ser Asp Leu Asn Lys Glu Asn Ser		
705	710	715 720
Arg Asn Thr Asn Ile Ser Ser Lys Ile Ala Ser Leu Val Arg Gln Lys		
725	730	735
Arg Glu His Glu Lys Lys Glu		
740		

<210> 223
<211> 397
<212> PRT
<213> *Saccharomyces* sp.

<400> 223
Met Leu His Gln Lys Ile Ala His Lys Val Arg Lys Val Val Val Pro
1 5 10 15
Gly Ile Ser Leu Leu Ile Phe Phe Gln Gly Cys Leu Ile Leu Leu Phe
20 25 30
Leu Gln Leu Thr Tyr Lys Thr Leu Tyr Cys Arg Asn Asp Ile Arg Lys
35 40 45
Gln Ile Gly Leu Asn Lys Thr Lys Arg Leu Phe Ile Val Leu Val Ser
50 55 60
Ser Ile Leu His Val Val Ala Pro Ser Ala Val Arg Ile Thr Thr Glu
65 70 75 80
Asn Ser Ser Val Pro Lys Gly Thr Phe Phe Leu Asp Leu Lys Lys Lys
85 90 95
Arg Ile Leu Ser His Leu Lys Ser Asn Ser Val Ala Ile Cys Asn His
100 105 110
Gln Ile Tyr Thr Asp Trp Ile Phe Leu Trp Trp Leu Ala Tyr Thr Ser
115 120 125
Asn Leu Gly Ala Asn Val Phe Ile Ile Leu Lys Lys Ser Leu Ala Ser
130 135 140
Ile Pro Ile Leu Gly Phe Gly Met Arg Asn Tyr Asn Phe Ile Phe Met
145 150 155 160
Ser Arg Lys Trp Ala Gln Asp Lys Ile Thr Leu Ser Asn Ser Leu Ala
165 170 175
Gly Leu Asp Ser Asn Ala Arg Gly Ala Gly Ser Leu Ala Gly Lys Ser
180 185 190
Pro Glu Arg Ile Thr Glu Glu Gly Glu Ser Ile Trp Asn Pro Glu Val
195 200 205
Ile Asp Pro Lys Gln Ile His Trp Pro Tyr Asn Leu Ile Leu Phe Pro
210 215 220
Glu Gly Thr Asn Leu Ser Ala Asp Thr Arg Gln Lys Ser Ala Lys Tyr
225 230 235 240
Ala Ala Lys Ile Gly Lys Lys Pro Phe Lys Asn Val Leu Leu Pro His
245 250 255
Ser Thr Gly Leu Arg Tyr Ser Leu Gln Lys Leu Lys Pro Ser Ile Glu
260 265 270
Ser Leu Tyr Asp Ile Thr Ile Gly Tyr Ser Gly Val Lys Gln Glu Glu
275 280 285
Tyr Gly Glu Leu Ile Tyr Gly Leu Lys Ser Ile Phe Leu Glu Gly Lys
290 295 300
Tyr Pro Lys Leu Val Asp Ile His Ile Arg Ala Phe Asp Val Lys Asp
305 310 315 320
Ile Pro Leu Glu Asp Glu Asn Glu Phe Ser Glu Trp Leu Tyr Lys Ile
325 330 335

Trp Ser Glu Lys Asp Ala Leu Met Glu Arg Tyr Tyr Ser Thr Gly Ser
 340 345 350

Phe Val Ser Asp Pro Glu Thr Asn His Ser Val Thr Asp Ser Phe Lys
 355 360 365

Ile Asn Arg Ile Glu Leu Thr Glu Val Leu Ile Leu Pro Thr Leu Thr
 370 375 380

Ile Ile Trp Leu Val Tyr Lys Leu Tyr Cys Phe Ile Phe
 385 390 395

<210> 224

<211> 303

<212> PRT

<213> *Saccharomyces* sp.

<400> 224

Met Ser Val Ile Gly Arg Phe Leu Tyr Tyr Leu Arg Ser Val Leu Val
 1 5 10 15

Val Leu Ala Leu Ala Gly Cys Gly Phe Tyr Gly Val Ile Ala Ser Ile
 20 25 30

Leu Cys Thr Leu Ile Gly Lys Gln His Leu Ala Gln Trp Ile Thr Ala
 35 40 45

Arg Cys Phe Tyr His Val Met Lys Leu Met Leu Gly Leu Asp Val Lys
 50 55 60

Val Val Gly Glu Glu Asn Leu Ala Lys Lys Pro Tyr Ile Met Ile Ala
 65 70 75 80

Asn His Gln Ser Thr Leu Asp Ile Phe Met Leu Gly Arg Ile Phe Pro
 85 90 95

Pro Gly Cys Thr Val Thr Ala Lys Lys Ser Leu Lys Tyr Val Pro Phe
 100 105 110

Leu Gly Trp Phe Met Ala Leu Ser Gly Thr Tyr Phe Leu Asp Arg Ser
 115 120 125

Lys Arg Gln Glu Ala Ile Asp Thr Leu Asn Lys Gly Leu Glu Asn Val
 130 135 140

Lys Lys Asn Lys Arg Ala Leu Trp Val Phe Pro Glu Gly Thr Arg Ser
 145 150 155 160

Tyr Thr Ser Glu Leu Thr Met Leu Pro Phe Lys Lys Gly Ala Phe His
 165 170 175

Leu Ala Gln Gln Gly Lys Ile Pro Ile Val Pro Val Val Val Ser Asn
 180 185 190

Thr Ser Thr Leu Val Ser Pro Lys Tyr Gly Val Phe Asn Arg Gly Cys
 195 200 205

Met Ile Val Arg Ile Leu Lys Pro Ile Ser Thr Glu Asn Leu Thr Lys
 210 215 220

Asp Lys Ile Gly Glu Phe Ala Glu Lys Val Arg Asp Gln Met Val Asp
 225 230 235 240

Thr Leu Lys Glu Ile Gly Tyr Ser Pro Ala Ile Asn Asp Thr Thr Leu
 245 250 255

Pro Pro Gln Ala Ile Glu Tyr Ala Ala Leu Gln His Asp Lys Lys Val
 260 265 270

Asn Lys Lys Ile Lys Asn Glu Pro Val Pro Ser Val Ser Ile Ser Asn
 275 280 285

Asp Val Asn Thr His Asn Glu Gly Ser Ser Val Lys Lys Met His
 290 295 300

<210> 225
<211> 1146
<212> DNA
<213> *Saccharomyces* sp.

<400> 225
atgtctttta gggatgtcct agaaagagga gatgaatttt tagaaggcta tcggcagaaga 60
agccccctt ggagattct ttcatcactg acatcattac tgaccccg ttgtatcaaaa 120
ctgccttcctt tcacatgcta taatgtcaaa ttgaatggg ttggaaaaatt agaaactgccc 180
ttggAACGTT cccaaaggaa aaatagggc cttatgacgg tcatgaacca tatgagtatg 240
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<220>
<223> Description of Artificial Sequence: Synthetic
Oligonucleotide

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Oligonucleotide

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<210> 235
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<223> Description of Artificial Sequence:Synthetic Oligonucleotide

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<210> 236

<211> 32

<212> DNA

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<211> 36

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<212> DNA

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<223> Description of Artificial Sequence:Synthetic Oligonucleotide

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International Bureau



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(10) International Publication Number
WO 00/18889 A3

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For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

WO 00/18889 A3

(54) Title: SEQUENCES OF PUTATIVE PLANT ACYLTRANSFERASES

(57) Abstract: By this invention, novel nucleic acid sequences encoding for acyltransferase related proteins are provided, wherein said acyltransferase-like protein is active in the transfer of a fatty acyl group from a fatty acyl donor to a fatty acyl acceptor. Also considered are amino acid and nucleic acid sequences obtainable from AT-like nucleic acid sequences and the use of such sequences to provide transgenic host cells capable of producing modified lipid content and composition.

INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 99/22231

A. CLASSIFICATION OF SUBJECT MATTER
 IPC 7 C12N9/10 C12N9/54 C12N9/82

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
 IPC 7 C12N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>NORBERG A. ET AL.: "Chemical detection of natural peptides by specific structures. Isolation chicken galanin by monitoring for its N-terminal dipeptide, and termination of the amino acid sequence." FEBS LETT 1991 AUG 19;288(1-2):151-3, XP000916139 abstract; figure 2</p> <p>---</p> <p>-/-</p>	1,9-18, 20

Further documents are listed in the continuation of box C.

Patent family members are listed in annex.

* Special categories of cited documents :

- "A" document defining the general state of the art which is not considered to be of particular relevance
- "E" earlier document but published on or after the international filing date
- "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- "O" document referring to an oral disclosure, use, exhibition or other means
- "P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search

7 July 2000

Date of mailing of the international search report

04.10.00

Name and mailing address of the ISA

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Meyer, W

INTERNATIONAL SEARCH REPORT

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PCT/US 99/22231

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	BROWN A P ET AL: "IDENTIFICATION OF A CDNA THAT ENCODES A 1-ACYL-SN-GLYCEROL-3-PHOSPHATE ACYLTRANSFERASE FROM LIMNANTHES DOUGLASII" PLANT MOLECULAR BIOLOGY, NL, NIJHOFF PUBLISHERS, DORDRECHT, vol. 29, no. 2, 1 October 1995 (1995-10-01), pages 267-278, XP002000905 ISSN: 0167-4412 abstract; figure 3	1 9-18,20
Y	ISHIZAKI O ET AL: "CLONING AND NUCLEOTIDE SEQUENCE OF COMPLEMENTARY DNA FOR THE PLASTID GLYCEROL-3-PHOSPHATE ACYLTRANSFERASE FROM SQUASH" FEBS (FEDERATION OF EUROPEAN BIOCHEMICAL SOCIETIES) LETTERS 1988, vol. 238, no. 2, 1988, pages 424-430, XP000916289 ISSN: 0014-5793 abstract; figure 2	1 9-18,20
Y	JOHNSON T C ET AL: "NUCLEOTIDE SEQUENCE OF ACYL-ACYL CARRIER PROTEIN GLYCEROL-3-PHOSPHATE ACYLTRANSFERASE FROM CUCUMBER" PLANT PHYSIOLOGY (BETHESDA) 1992, vol. 99, no. 2, 1992, pages 771-772, XP000919121 ISSN: 0032-0889 abstract	1 9-18,20
Y	LASSNER M W ET AL: "LYSOPHOSPHATIDIC ACID ACYLTRANSFERASE FROM MEADOWFOAM MEDIATES INSERTION OF ERUCIC ACID AT THE SN-2 POSITION OF TRIACYLGLYCEROL INTRANSGENIC RAPESEED OIL" PLANT PHYSIOLOGY, US, AMERICAN SOCIETY OF PLANT PHYSIOLOGISTS, ROCKVILLE, MD, vol. 109, no. 4, 1 January 1995 (1995-01-01), pages 1389-1394, XP002027767 ISSN: 0032-0889 abstract; figure 1	1 9-18,20
X	NAGIEC, M. MAREK ET AL: "A suppressor gene that enables <i>Saccharomyces cerevisiae</i> to grow without making sphingolipids encodes a protein that resembles an <i>Escherichia coli</i> fatty acyltransferase" J. BIOL. CHEM. (1993), 268(29), 22156-63, XP000644683 abstract; figure 2	9-18,20
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INTERNATIONAL SEARCH REPORT

Inte... pnal Application No
PCT/US 99/22231

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	NISHIDA I. ET AL.: "The gene and the RNA for the precursor to the plastid-located glycerol-3-phosphate acyltransferase of <i>Arabidopsis thaliana</i> ." PLANT MOL BIOL 1993 JAN;21(2):267-77, XP000916240 abstract; figure 2 ---	1
X		9-18,20
Y	WO 96 24674 A (GENE SHEARS PTY LTD ;SLABAS ANTONI RYSZARD (GB); BROWN ADRIAN PAUL) 15 August 1996 (1996-08-15) abstract; figure 1 ---	1
X		9-18,20
A	YOKOI SHUJI ET AL: "Introduction of the cDNA for <i>Arabidopsis</i> glycerol-3-phosphate acyltransferase (GPAT) confers unsaturation of fatty acids and chilling tolerance of photosynthesis on rice." MOLECULAR BREEDING JUNE, 1998, vol. 4, no. 3, June 1998 (1998-06), pages 269-275, XP000909905 ISSN: 1380-3743 abstract -----	1
X		9-18,20

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Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. Claims Nos.: because they relate to subject matter not required to be searched by this Authority, namely:

2. Claims Nos.: because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

3. Claims Nos.: because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

See additional sheet

1. As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

1, partially 9-18, 20, 21

Remark on Protest

- The additional search fees were accompanied by the applicant's protest.
- No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA 210

1. Claims: 1, partially 9-18, 20, 21
relating to Seq Id No 127
2. Claims: 2, partially 9-18, 20, 21
relating to Seq Id No 128
3. Claims: 3, partially 9-18, 20, 21
relating to Seq Id No 129
4. Claims: 4, partially 9-18, 20, 21
relating to Seq Id No 132
5. Claims: 5, partially 9-18, 20, 21
relating to Seq Id No 130
6. Claims: 6, partially 9-18, 20, 21
relating to Seq Id No 133
7. Claims: 7, partially 9-18, 20, 21
relating to Seq Id No 131
8. Claims: 8, partially 9-18, 20, 21
relating to Seq Id No 134
9. Claims: partially 9-18, 20, 21, 22
relating to Seq Id No 1
10. Claims: partially 9-18, 20, 21, 22
relating to Seq Id No 10
11. Claims: partially 9-18, 20, 21, 22
relating to Seq Id No 12

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

12. Claims: partially 9-18, 20, 21, 22

relating to Seq Id No 14

13. Claims: partially 9-18, 20, 21, 22

relating to Seq Id No 16

14. Claims: partially 9-18, 20, 21, 23

relating to Seq Id No 3

15. Claims: partially 9-18, 20, 21, 22

relating to Seq Id No 5

16. Claims: partially 9-18, 20, 21, 22

relating to Seq Id No 7

17. Claims: partially 9-18, 20, 21, 22

relating to Seq Id No 18

18. Claims: Invention No. 18-126: Claims 9-22 all partially

each individual invention relating to Seq Id No. 24 to Seq
Id. 126, respectively

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/US 99/22231

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
WO 9624674 A	15-08-1996	AU CA CA EP	4669096 A 2212570 A 2235267 A 0808368 A	27-08-1996 15-08-1996 24-04-1997 26-11-1997
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